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Soybean Research Report

2018

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(Alabama A&M University and Auburn University)

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Confidential Report

Contents

I. Cultural Management

Cover Crop Mixtures for Soybean Production

D. Delaney, K Balkcom, A. Price, Y. Feng, and A. Gamble

Introduction:

Alabama row crop producers have widely adopted conservation tillage, with many using winter cover crops before summer crops, including soybeans. Recently, cover crop seed mixtures have been promoted and adopted by producers in soybean growing areas. There is little data available in the southeast U.S. to show whether there is an advantage for soybean producers to use cover crop mixtures compared to a single species, and how to manage planting them, including the proper ratio of each seed component.

Procedures:

Field studies were initiated at two Experiment Station locations (EV Smith = EVS and Prattville = PEF) using different combinations and ratios of cover crops, beginning in the fall of 2015 for the 2016 soybean crop. The same treatments of cover crops were planted again in the fall of 2016, 2017 and 2018. Twelve cover crop treatments were tested, with one to three species (cereal rye, crimson clover, and radish) with varying seed ratios of each, such as full, one-half or one-third of the recommended rates. Each treatment was replicated 4 times. The EVS site was in a 2-year rotation with cotton and the same treatments were repeated on the plots each year, so that 2019 will be the 4th cash cropping year, or two complete cropping rotations.

A similar study using nine cover crop treatments was planted at the Black Belt REC in the fall of 2018, with a replicated study for soybean yield on a Vaiden soil ("normal pH"), and another strip trial to study survival and biomass production on a high pH Sumter soil. Species used included rye, Cosaque oat, wheat, radish, radish-turnip hybrid, crimson clover, and Austrian winter peas as well as 2-way and 3-way mixture treatments.

Cover crops biomass samples of 2 * ¼ meter^2 per plot were taken on 20 April 2018 before termination, dried, weighed, and analyzed for carbon and nitrogen content. Soybeans were planted no-till approximately 2 weeks later, after covers had dried down. Measurements were also made of soybean stand counts, height and yield.

Results:

Good growth of cover crops was made, especially of those treatments including rye and/or crimson clover (Table 1). Wet weather in spring to early summer delayed planting of soybean at EV Smith and Prattville. Packing rains and wet conditions led to less than optimum soybean stands at both locations, ~ 120,000 plants/A at EVS and 85,000 plants/A at PEF. Some stand differences between treatments were noted at EVS but not at PEF (data not shown). There were no significant differences in soybean plant height at EVS, while at PEF several treatments were taller than the Fallow. At EVS, there were no significant differences between treatments for yield in 2018. However, treatment 4: Radish @ 8 lb/A and treatment 7: Rye @ 45 lb/A + Radish @ 8 lb/A yielded less than the Fallow treatment. (Table 1).

Table 1. Biomass, Soybean Height and Yield Following Cover Crop Mixtures at EVS and PEF

	Seeding rates			**Biomass**		**Soy height**		**Yield**	
	Lb/A			**Lb/A**		**inches**		**Bu/A**	
Treatment	Rye	Cr Clover	Radish	EVS	PEF	EVS	PEF	EVS	PEF
1-Fallow	0	0	0	201	781	33.2	27.4	49.0	50.6
2	90	-	-	6020	3167	35.0	29.0	55.5	-
3	-	20	-	3879	4595	34.1	29.8	45.8	47.6
4	-	-	8	4236	1428	34.3	26.4	47.3	44.7
5	45	20	-	4437	4773	34.3	27.8	54.3	45.1
6	30	20	-	3701	4885	33.4	28.3	48.3	49.2
7	45	-	8	4459	2119	34.3	27.8	50.7	44.1
8	30	-	8	3991	2097	32.1	27.5	44.2	47.0
9	-	20	8	3344	4305	35.6	27.7	51.7	46.3
10	-	10	8	4147	4372	34.1	29.8	44.9	47.1
11	45	10	4	3701	5532	35.1	29.3	47.4	50.1
12	30	10	4	5485	3859	32.9	28.1	52.1	45.4
		LSD *(p=0.10)*		*1285*	*1224*	*NS*	*1.57*	*NS*	*5.22*

Soil Quality

Plots were also sampled at EVS for soil quality parameters including organic carbon, microbial biomass, arbuscular mycorrhizal fungi (AMF), active Carbon, soil respiration and glomalin-related soil protein (GRSP). In the first 2 years of the study, the only significant differences were for AMF colonization of cotton roots. However, in the 3rd year, several differences were noted for the soil factors studied, showing the time needed for detectable changes in soil quality.

No significant differences were noted for soil organic carbon (organic matter), while the active carbon fraction was significantly greater for rye alone, rye/clover mixture, and rye/radish mix treatments compared to fallow by the 3rd year. However, 3-way and clover/radish mix treatments were not significantly higher than fallow for active carbon.

Soil respiration as an indicator of microbial activity was higher for cover crop treatments than fallow in 2018, and was much higher for the rye/clover mixture.

AMF are beneficial fungi associated with plant roots, stimulating plant growth and improving physical qualities of the soil. Radish, as a member of the Brassica family, is not known to support AMF and has been known to decrease AMF in following crops. However, this experiment showed that including radish in a mixture with rye or clover did not decrease AMF colonization, but those mixtures still increased AMF compared to fallow plots.

Overall, little advantage was noted for the 3-way mixtures compared to 2-way (rye/clover) or single species (rye) for soil health measurements.

Soybean Production Tools for Alabama

D. Delaney, E. Sikora, K. S. Lawrence, M. Runge, R. Yates, E. McGriff, C. Hicks, K. Wilkins, B. A. Dillard, and T. Sandlin

Objectives:

Objective 1: To evaluate soybean cultivars suitable for Alabama growing conditions under producer practices and growing conditions.

Results: On-farm variety trials were planned at four sites across the state with seed obtained from participating seed companies and cooperators lined up. The wet spring caused delays for many of our planned farmer-cooperators, so that planting of some trials was much delayed, and other potential cooperators switched to other crops or declined to participate. Of the 4 trials planned, only 3 could be planted and harvested: in Marengo, Jackson and Montgomery Counties. All locations were dryland. The fourth location planned was an Irrigated MG 4.

In Jackson County, an early season MG 5/late MG 4 dryland trial was planted on 08 May with 13 varieties and harvested 08 October. Yields ranged from 30 to 40 bu/A, with a test average of 35.4 bu/A.

In Marengo County, a MG 5/late MG4 dryland trial was planted on 09 June in a Black Belt soil with a pH of 7.8. Iron chlorosis (deficiency) was moderate due to regular rainfall, but was enough to be rated by late summer. Chlorosis ranged from 1 (no chlorosis) to 5 on a visual scale (10 = plant death). Plots were harvested on 19 October with yields ranging from 26 to 48 bu/A.

In Montgomery County, a MG 5/6 dryland trial with 11 varieties was planted on 11 June and harvested 24 October. Yields were good for this sandy loam soil and the 10 varieties harvested ranged from 45 to 54 bu/A, with a test average of 49.9 bu/A. (Deer damage on the variety nearest a tree line rendered data from it unreliable).

Objective 2: Evaluate the use of treatments to control iron chlorosis on high pH Black Belt soils.

Results: Some reports as well as field observations have shown that increased populations of soybeans can lessen symptoms of iron chlorosis on high pH soils. An experiment was planted at the Black Belt REC on a high pH (8.1) soil using six seeding rates ranging from 90,000 to 240,000 seeds/acre of a variety rated relatively tolerant for iron chlorosis. With plentiful rainfall during the summer of 2018, little chlorosis was present and was not enough for an accurate rating. However, yields were still affected by planting rate, ranging from 34 to 38 bu/A. (Table 1.)

Table 1. Soybean Seeding Rates for high pH (8.1) Soils, Black Belt REC 2018

Treatment #	Seeding rate/A	Yield bu/A
1	90,000	34.3
2	120,000	38.4
3	150,000	37.1
4	180,000	37.4
5	210,000	33.0
6	240,000	36.0
	LSD (p=0.10)	*2.94*

Objective 3: Evaluate nitrogen applications to soybeans in high yield environments. Several soybean yield contest winners across the country have applied additional N fertilizer for yield enhancement; however, we have limited data in AL at high yield levels whether N application has benefits, while optimum timing and rates are unknown.

Results: Tests were conducted at the Tennessee Valley REC, EV Smith Field Crops Unit and Sand Mountain REC with soybeans under irrigation. Planting was delayed at most sites due to wet soils. At TVREC, Pioneer 55A49X was planted on 22 May in 30-inch rows. At EVS, Pioneer 67T90R2 was planted on 05 June in 36-inch rows, and planting was made at SMREC ~ 18 May in 30-inch rows. Urea nitrogen was applied either At-plant, at the R3 growth stage, or split between the R3 stage and 21 days later. Gypsum was used to supply 20 lb/A of sulfate-sulfur to all plots. Rates of 0, 40, 80, 120 or 160 lb/A of N were used. Plots were monitored for lodging and other problems, while seed samples were analyzed for 100-seed weight and protein and oil content (data not shown).

At TVREC, yields ranged from 80.9 to 85.6 bu/A with no statistically significant effects on yield for the Rate of N applied (Table 2). At EVS, yields ranged from 32.3 to 40.4 bu/A with no significant differences. At SMREC yields ranged from 51.7 to 59.8 bu/A with no significant yield differences for rate of N applied.

No effects on plant height, lodging or delayed maturity were noted in any trial. Data analysis is ongoing to determine effects of timing or splitting of N application on seed quality. At TVREC, higher rates of N @ R3+ decreased protein compared to the Untreated check in 2018.

Table 2. Effect of Nitrogen Application Rates and Timing for Soybeans, TVREC, EVS-FCU, and SMREC 2018

Nitrogen	**Nitrogen**	**TVREC**	**EVS-FCU**	**SMREC**
Timing	**Rate (lb/A)**	**Bu/A**	**Bu/A**	**Bu/A**
At-plant	40	83.3	34.3	51.7
At-plant	80	84.9	39.5	55.5
At-plant	120	84.5	37.9	58.1
At-plant	160	85.6	33.9	57.4
R3	40	84.8	32.3	53.2
R3	80	81.6	35.9	57.7
R3	120	82.7	38.6	59.8
R3	160	80.9	40.4	58.3
Split: R3, + 21 days	80	82.3	33.7	53.5
Split: R3, + 21 days	120	84.5	34.2	54.4
Split: R3, + 21 days	160	81.1	35.5	57.1
Untreated	0	83.1	33.8	54.0
	LSD (p=0.10)	*NS*	*NS*	*NS*

Continued Support of Long Term Research

D. Delaney, K. Balkcom, A. Gamble

Rationale

The "Old Rotation" (c. 1896) at Auburn is the oldest, continuous cotton experiment in the world. It consists of 13 plots on 1 acre. Treatments include with and without winter legumes, timing of fertilizer application, 2-year rotations with corn with and without winter legumes, and a 3-year rotation with corn, wheat and soybeans. In 2003, plots were split and irrigation was installed on half of the plots.

The Cullars Rotation (c. 1911) at Auburn is a 3-year rotation of cotton (crimson clover/vetch)-corn (wheat)-soybeans with soil fertility variables on approximately 3 acres of land. This is the oldest soil fertility experiment in the South and has 14 soil fertility treatments replicated 3 times. It was placed on the National Register in April 2003. This experiment is highly visible because of its location adjacent to the Jules Collins Smith Museum of Art in Auburn. It occupies the site where cotton rust was first associated with a potassium deficiency.

Experimental Methods

Experiments continued with long-term treatments applied and managed according to modern recommended practices, data recorded and summarized, and papers presented at state, regional, and national meetings. The Long-term Crop Rotations continued to be available for AU Student Special Projects, research by other Universities in Alabama and other states, for field labs by classes, and for numerous campus visitors.

Report

The long-term rotations at Auburn University continue to be invaluable "Outdoor Classrooms" for students and visitors to Auburn University. During the 2018 season, at least ten tours were given of the Old Rotation and/or the Cullars Rotation. Tours included two AGRI1000 (Introduction to Agriculture) classes, four CSES1000 (Basic Crop Science) classes, an Extension county agent group from Texas, the Southern Cover Crops Council, Yara interns, and a group of 5th and 6th graders as part of a Science

Discovery Camp. Improved signage has been ordered to increase visibility of the Old Rotation and Cullars Rotation, which will be installed in 2019.

The Cullars Rotation is a valuable experiment to teach students visual symptoms of various nutrient deficiencies (Photo 1). The effect of poor fertility and lime management is clearly demonstrated through soybean yield data (Table 1).

Table 1. Average soybean yields according to treatment for the Cullars rotation.

Fertility Treatment	Yield (bu/acre)
No N, winter legume	45.7
No N, no winter legume	45.4
No fertility since 1911	0.1
Complete fertility, no legume	45.0
No P	26.9
Complete N-P-K, no micros	40.0
4/3 K rate	37.1
Rock phosphate	47.8
No K	19.2
2/3 K Rate	36.7
No lime (pH ~4.5)	0.6
No S	46.0
Complete fertility	41.9
1/3 K rate	38.6

The Old Rotation continues to demonstrate the benefits of crop rotation with soybean, wheat, corn, and winter cover crops to sustainable cotton production in dryland and irrigated cotton production systems in the Southeast. Rotations without legume cover crops remain stagnant even with improved varieties and technology, while rotations including a winter legume continue to improve even without additional N. For the 2018 growing season, lint yields for continuous cotton with no crop rotation, no supplemental N, and no winter legume averaged 640 lbs per acre. Lint yields for

cotton rotated with corn and a winter legume, still without supplemental N, averaged 1980 lbs per acre. Organic matter has nearly doubled for treatments with high residue inputs (i.e. rotations with soybean, corn, wheat or winter legume cover crops) when compared to continuous cotton with no cover crop and no N applied (Table 2), leading to increased yield potential.

Photo 1. The Cullars Rotation is a valuable demonstration site to teach students symptoms of nutrient deficiencies in soybean, such as the early-season potassium deficiency observed in this photo.

Table 2. Average cotton lint yields according to treatment for the Old Rotation. Results demonstrate the benefits of crop rotation with corn, wheat, soybean, and winter legume cover crops.

Treatment				2007-2017 Avg. Cotton Lint Yield	
Rotation	Winter Cover Crop	N Fertilizer (120 lbs/A)	Organic Matter	Dryland	Irrigated
			-- % --	-- lb/acre--	– lb/acre –
Continuous Cotton	No	No	1.6	476	589
Continuous Cotton	**Yes**	**No**	**2.9**	**1118**	**1331**
Continuous Cotton	No	Yes	2.7	1057	1407
Cotton-Corn	**Yes**	**No**	**2.6**	**1305**	**1468**
Cotton-Corn	Yes	Yes	2.9	1290	1680
Cotton-Corn-Wheat-Soybean	**Yes**	**Yes**	**3.1**	**934**	**1255**

Irrigation Strategies for Alabama Black Belt Soils

T. Knappenberger, J. Shaw, and E. Brantley

Introduction

Several soybean growers in Alabama's Black Belt Region have installed central pivot irriga- tion systems in recent years. A 2015-2017 project (Improvement of Irrigation Management on Alabama Black Belt Soils) has shown that irrigation increases the yield in soybeans. The Black Belt clayey soils are characterized by low infiltration rates; shrink-swell behavior, and pronounced surface cracking. For an effective irrigation strategy and to avoid surface water runoff it is necessary to determine maximum infiltration rates and cracking behavior of the clays in the Black Belt region.

Material & Methods

Soil Characterization

Soils were sampled in different locations across the blackbelt. Disturbed samples were ana- lyzed for soil texture, soil carbon content, and the coefficient of linear extensibility (COLE)– a measure for the shrink and swelling behavior of a soil. Undisturbed samples were also collected to measure the hydraulic conductivity and the water retention curve.

Runoff Experiments

The runoff experiments were performed in April 2018. A runoff simulator was constructed by fastening a spray nozzle on top of a 10 foot high tripod. 0.25 inch steel plates were driven into the soil to form a one square meter area. Downslope of that area the soil was excavated and a gutter and rain gauge was installed in a way that the gutter collects the runoff from the test plot and the rain gauge measures the flow. Each runoff experiment was performed for 15 minutes.

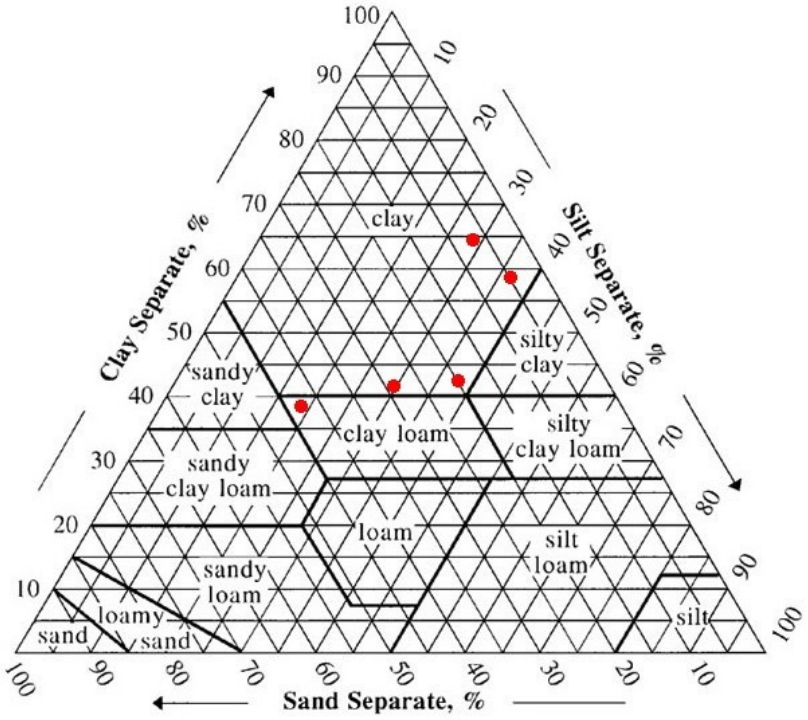


Figure 1: Texture of the tested blackbelt soils.

Results

Soil Characterization

All soil samples were classified as clay or clay loam (Table 1, Figure 1). The carbon content was above average for Alabama. the coefficient of linear extensibility (COLE) was between 13.2 and 20.4%. This means that in the tested soils cracks can make up a volume of up to 20.4%. While the USDA texture classes and triangles are standard for agricultural appli- cations this system fails to recognized different clays and clay properties. The Unified Soil Classification System (USCS) is more widely used in engineering and geophysical applica- tions. The main difference is that in the USCS the plasticity of a soil is also determined. The Atterberg limits are an important part of the USCS and they include the liquid and plastic limit. Those limits indicate the water content of a soil when it transitions from solid to plastic (plastic limit) and from plastic to liquid (liquid limit). Liquid limits were high and all soils other than the Vaiden Clay classified as fat clays in the Unified Soil Classification System (USCS).

Table 1: Soil characteristics for the processed samples with the coefficient of linear extensi- bility (COLE), liquid limit (LL), plastic limit (PL), and plasticity index (PI).

Soil	**Sand** (%)	**Silt** (%)	**Clay** (%)	**Texture** (-)	**Carbon** (%)	**COLE** (%)	**LL** (g/g)	**PL** (g/g)	**PI** (-)
Houston Clay	6	36	58	Clay	2.6	19.2	61.6	26.6	35.0
Sumter Silty Clay	21	37	42	Clay	8.7	13.2	50.2	27.0	23.2
Vaiden Clay	44	18	38	Clay Loam	2.7	14.9	47.1	30.0	17.1
Oktibbeha Clay	30	29	41	Clay	2.7	20.4	50.7	22.0	28.7
Leeper Silty Clay	8	28	64	Clay	2.7	18.3	65.0	24.0	41.0

Hydraulic Conductivity

The saturated hydraulic conductivity K_s was measured in the lab utilizing the undisturbed soil samples. Usually, a K_s measurement reaches a steady state within hours and stays stable at that rate. For all tested blackbelt soils we experienced an interesting pattern. The soils were tested at field moisture which was the first saturated hydraulic conductivity measurement and typically it was higher than 1,000 cm/day (400 in/day) (Figure 2).

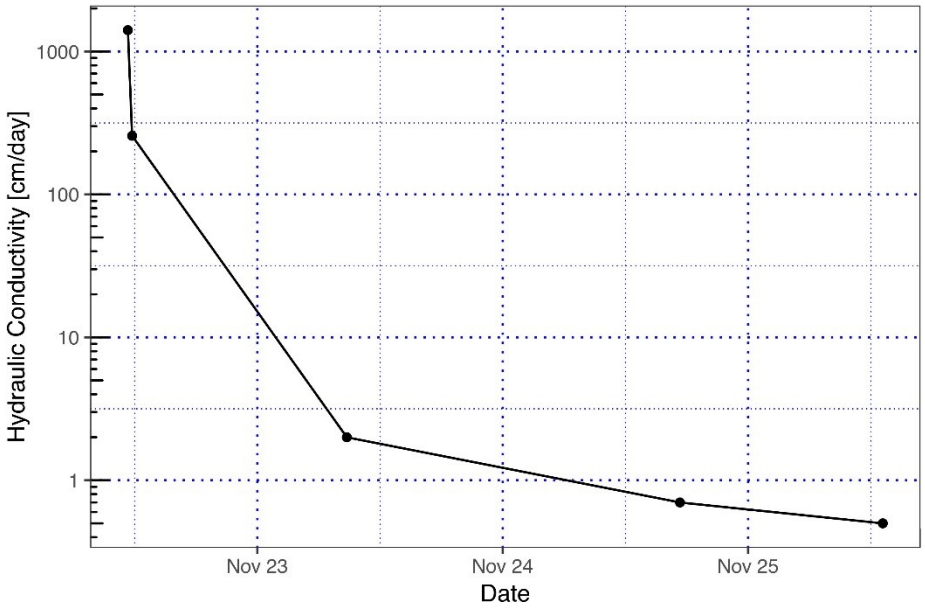


Figure 2: Evolution of the hydraulic conductivity of a Leeper Clay over time.

As we continued the measurement over the next days the saturated hydraulic conduc- tivity would significantly drop and usually would converge to a value less than 1 cm/day (0.4 in/day). We have seen this pattern in all tested soils. This is a saturated hydraulic conductivity dynamic we were not aware of and we think is worth further exploration. Ap- parently, there are smaller cracks in the soil samples which allow high saturated hydraulic conductivity measurements. Interestingly, these

cracks take a few days to close which means the saturated hydraulic conductivity remains relatively high. In terms of irrigation this means that the soil would not readily limit infiltration.

If it takes days for cracks to close irrigated soils may not even experience closed cracks. A fully developed crop can transpire up to 1 cm (0.4 in) per day. Assuming that 2.5 cm (1 in) of water was irrigated the crop could consume this additional water within three days which would be before the cracks completely close. A crack free soil during the growing season is therefore only likely of rainfall persists over a few days.

Runoff Experiments

Figure 3: This image illustrates the runoff experimental setup with the spray nozzle on the tripod and the one square meter area draining into the gutter.

Figure 3 shows the rainfall simulator and the experimental plot. For the runoff experi- ments we applied 2 cm of irrigation for a 15 min period which is equivalent to an irrigation rate of 200 cm/day and therefore quite high. Irrigation water started to runoff minutes after the experiments were started. We tested bare soils and soils with rye and ryegrass cover crop. The soils with cover crop had less runoff (Figure 4). In the cover crop plots the irrigation water was intercepted and also temporarily stored on the canopy which means less irrigation water hit the soil in the first place in comparison to the bare soil experiments. And second in the cover crop experiments the rye and ryegrass maintain an active root system which increases infiltration.

Interception of irrigation water is an important mechanism also relevant when deter- mining the irrigation depth and irrigation efficiency. The literature relates interception to the leaf area index. A full canopy crop can easily store 0.6 cm (0.25 in) of irrigation water which means that amount never reaches the soil surface and does not contribute to pant growth.

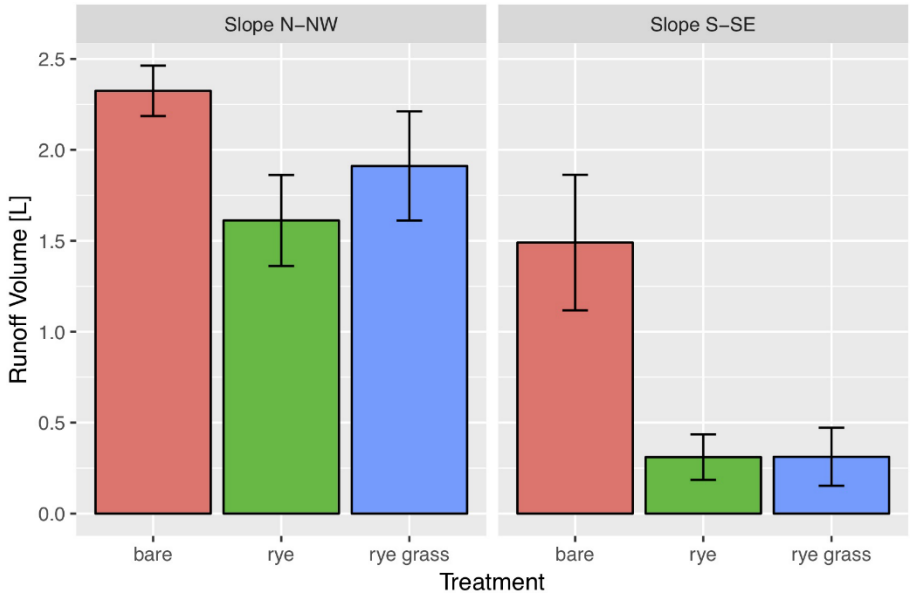


Figure 4: Runoff on bare soil was higher than on soils planted with cover crop.

Variable Rate Irrigation Based on Soil Sampling and Sensor Techniques

T. Knappenberger, A. Poncet, D. Monks, and G. Pate

Introduction

Recent advances in agriculture provided a better understanding of crop response to field management strategies and in-field spatial variability. Technologies are being developed to allow more precise placement of seeds, water, and other inputs. It is now possible to adapt input placement to yield goals and these techniques will become more valuable as the technology advances. The Alabama Agricultural Experimental Station has made large investments in Variable-Rate Irrigation (VRI) and a field-scale, variable-rate center pivot has been in use at the E.V. Smith Research Center in Shorter, AL (EVSREC) since 2013. Greg Pate, EVSREC director, has been working with the College of Agriculture and several Faculty to conduct research that will make full use of this technology. This study was conducted in 2018 to assess soybean response to different irrigation strategies.

Material & Methods

The cultivars Pioneer 52A26R *(Cultivar 1)*, Pioneer 55T81R *(Cultivar 2)*, and Asgrow 5831RR *(Cultivar 3)* were evaluated for yield response to the following irrigation strategies:

- **Rainfed**: no-irrigation;
- **Checkbook**: irrigation was applied throughout the growing season (from V2 to R6). Water deficit in the root zone was estimated on a daily basis using soil and weather data. Crop evapotranspiration was estimated as a function of growth stage.
- **Sensor-Based**: irrigation was applied throughout the growing season (from V2 to R6). Water deficit in the root zone was measured using watermark sensors placed at 8, 16, and 24 in depth in the soil profile. Irrigation in these plots was triggered when measured soil matric potential was lower than -50 kPa.
- **From R3 to R4, R4 to R6, and R3 to R6**: irrigation was applied only in the reproductive stage during (respectively): pod formation, seed filling, and both pod formation and seed filling. Water deficit in the root zone was measured using watermark sensors placed at 8, 16, and 24 in depth in the soil profile. Irrigation in these plots was triggered when measured soil matric potential was lower than -50 kPa.

Cultivar and irrigation treatments were layout under the variable-rate irrigation pivot as presented in Figure 1. Irrigation decision was made bi-weekly. Cumulative irrigation depth applied to each cultivar x irrigation treatments were summarized in Table 1.

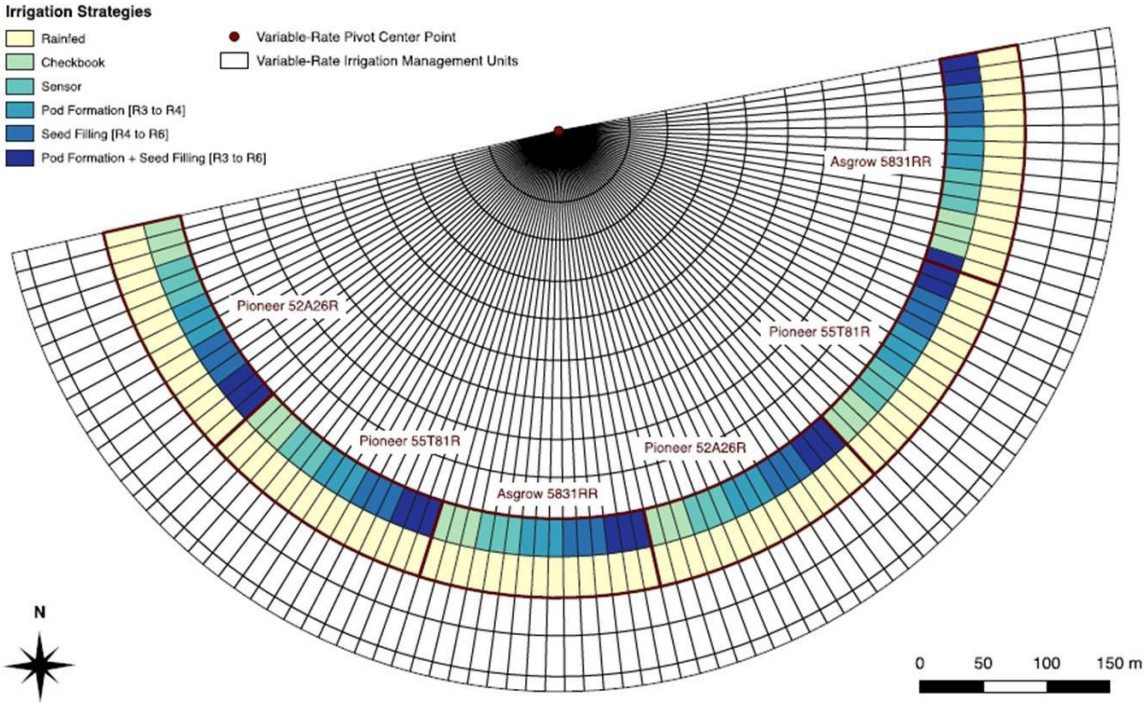


Figure 1: Treatment layout.

Table 1: Cumulative irrigation depths [in] applied to by irrigation treatment and cultivar.

Irrigation Stragegy	Cultivar 1	Cultivar 2	Cultivar 3
Checkbook	(4.0/4.2)	(4.2/4.2)	(4.2/4.2)
Sensor	(2.4/2.4)	(1.8/2.5)	(2.1/1.9)
R3-R4	(1.0/0.9)	(0.9/1.1)	(1.2/0.9)
R4-R6	(1.9/0.5)	(1.4/1.3)	(1.5/0.9)
R3-R6	(1.6/2.1)	(1.1/1.6)	(1.8/2.8)

All cultivars were planted on May 15, 2018 and harvested on November 21, 2018. Seeding rate was 140,000 seeds/ac. A normal year has a monthly average air temperature and rainfall that is within plus or minus one standard deviation of the last 20 years mean monthly average air temperature and rainfall. The first half of 2018 was comparable to the normal year (Figures 2 and 3). The second half of 2018 was overall warmer and wetter than the normal year.

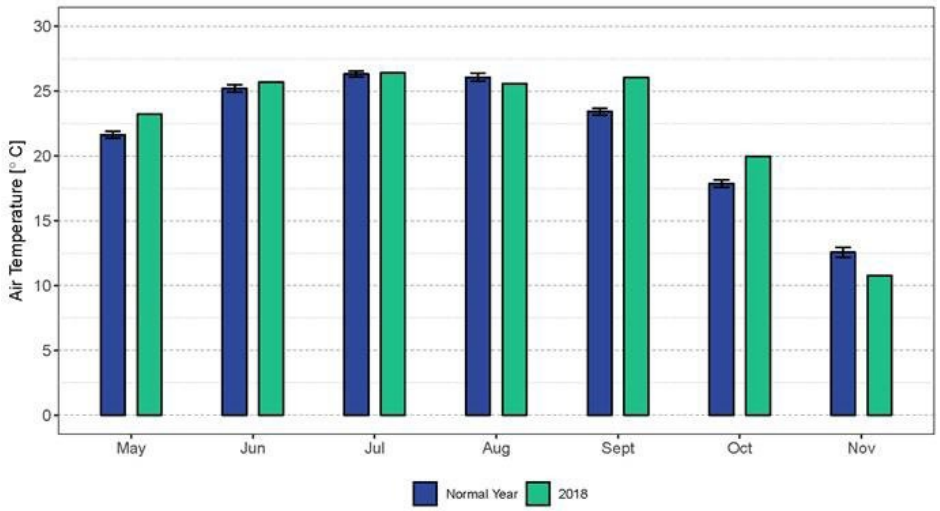


Figure 2: Monthly average air temperature in 2018 and comparison to historical data.

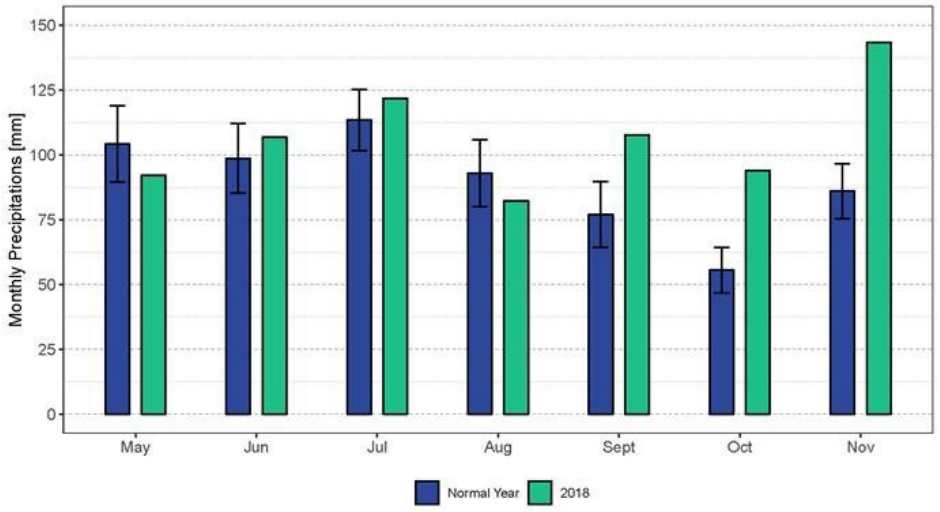


Figure 3: Monthly rainfall in 2018 and comparison to historical data.

Data were collected along two spatial transects made of 300 equidistant sites (Figure 4). Soybean vegetative development was evaluated on a weekly basis throughout the growing season by measuring Leaf Area Index (LAI). Soybean reproductive development was mea- sured at maturity through measurements of pod and seed weights. Yield was measured at harvest using a yield monitor.

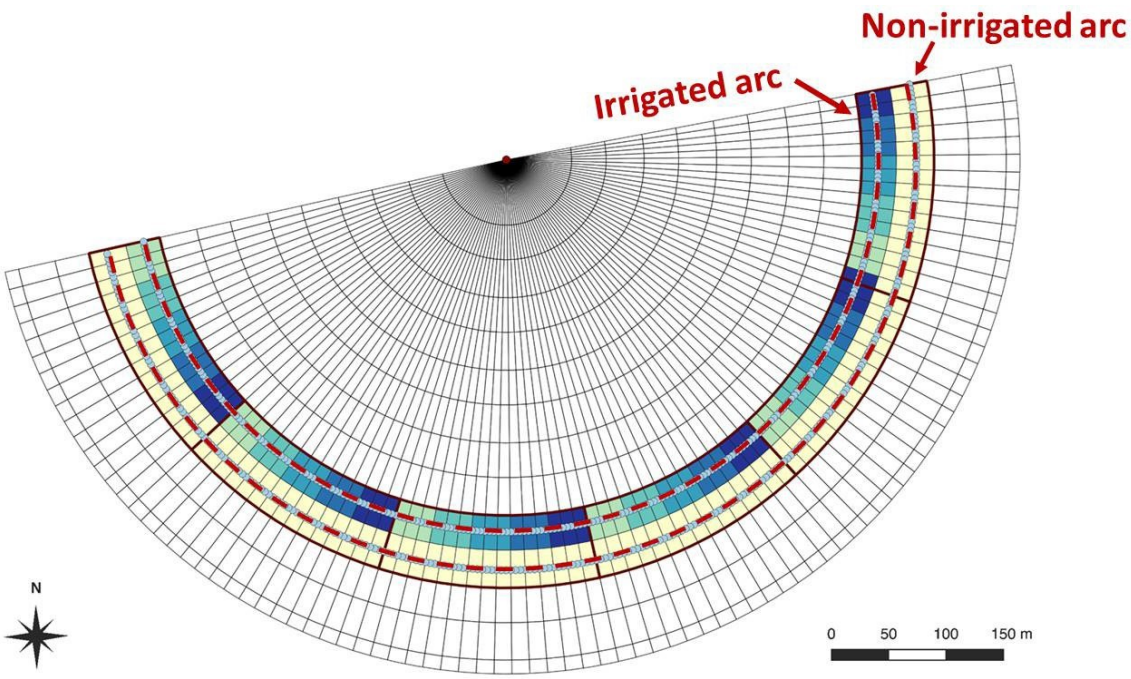


Figure 4: Transects used for data collection.

Results

Yield

Cultivar 3 maximized yield in the irrigated arc. Cultivar 2 maximized yield in the non- irrigated arc. Stronger differences between cultivars were observed in the irrigated arc than in the non-irrigated arc (Figure 5). Irrigating during seed filling maximized yield for culti- var 2 and 3. The sensor strategy maximized yield for cultivar 1. Stronger irrigation effects were observed with cultivars 2 and 3 than with cultivar 1 (Figure 6). Statistical analysis demonstrated that only 16% and 47% of yield variability was explained by the treatment effects in the irrigated and non-irrigated arcs, respectively. The rest was explained by the site-specific effects occuring within the field.

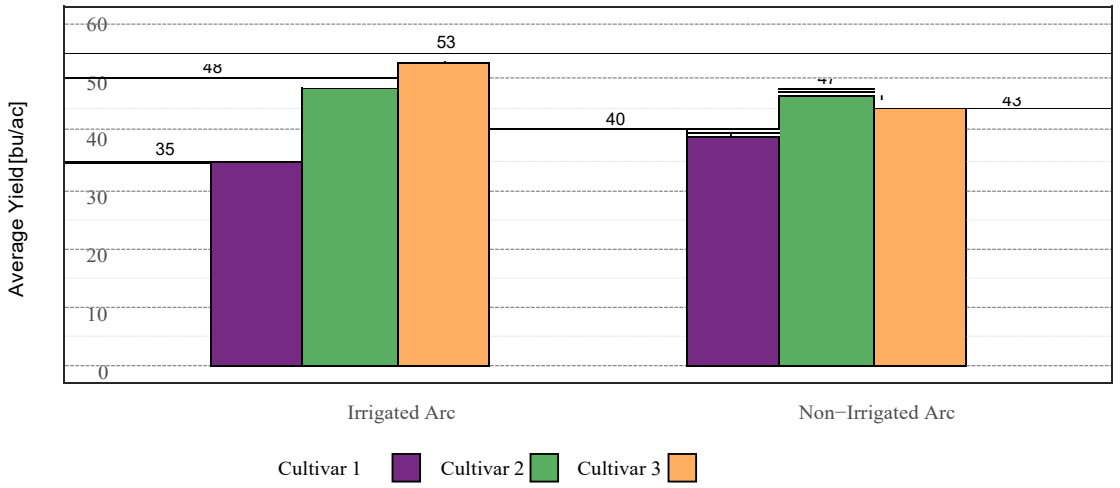


Figure 5: Average yield by cultivar in the irrigated and non-irrigated arcs. Error bars indicate the standard error observed for each cultivar across the spatial transects.

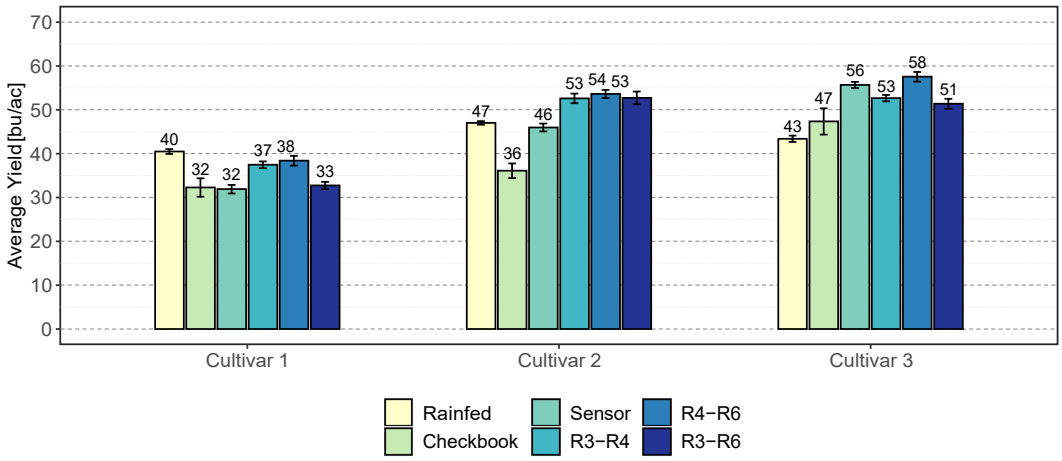


Figure 6: Average yield by cultivar in the irrigated and non-irrigated arcs. Error bars indicate the standard error observed for each cultivar x irrigation treatment across the spatial transect.

Soybean Growth

Cultivars 2 and 3 were two determinate varieties: the soybeans grew to their final height before flowering, all nodes flowered simultanously, and pod development was uniform along the plant. Cultivar 1 grew as an indeterminate variety: soybeans flowered early in the growing season, the lower nodes flowered first, and the lower pods developped earlier than the upper pods on along the plants. This explained why cultivar 1 entered in the reproductive stage two to three weeks before cultivars 2 and 3. All three cultivars achieved maturity at approximately the same time.

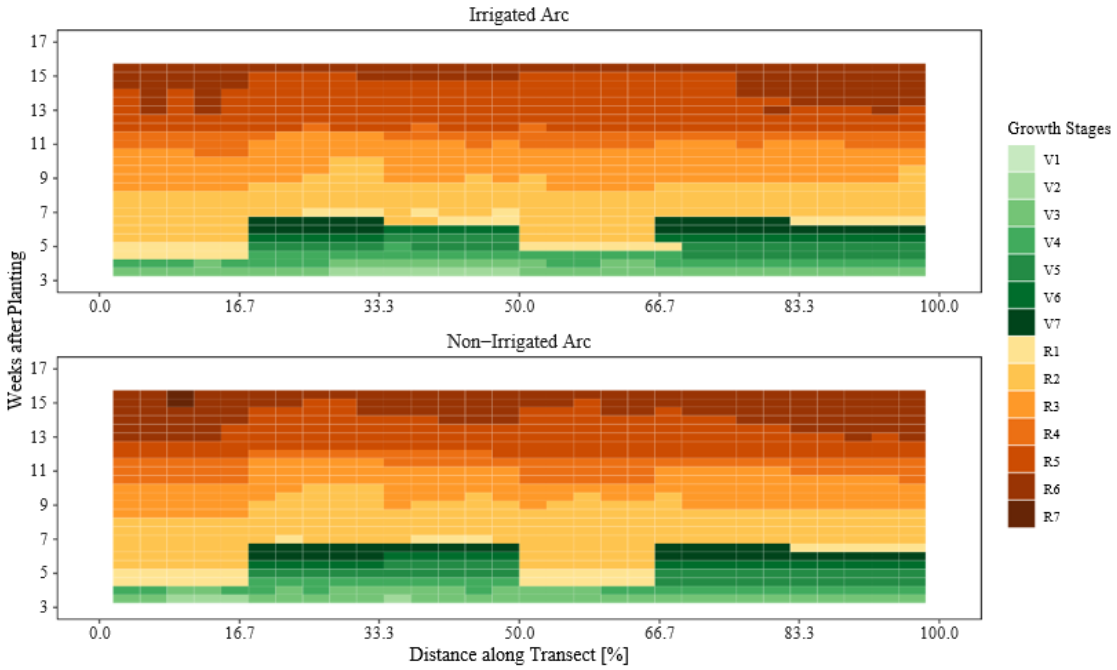


Figure 7: Summary of soybean growth along the irrigated and non-irrigated arcs.

Reproductive Development

The following data were collected along the spatial transect to characterize soybean repro- ductive development: pod weight per plant, 100 seed weight, and number of seeds per pod. The weight of seeds per pod and the number of pods per plant were then estimated using equations (1) and (2)

$$SeedWeightperPod = \frac{100\ \text{Seed Weight}}{100} \cdot \text{Number of Seeds per Pod} \quad (1)$$

$$DruNumberofPods = \frac{\text{Dry Pod Weight per Plant} \cdot 0.7}{\text{Seed Weight in Pod}} \quad (2)$$

6

Strong positive correlations were identified between the estimated weight of seeds in a pod and the number of pods per plant (Figure 8). Overall, cultivar 1 and 2 produced smaller seeds and less pods than average, with little differences between treatments and limited variability across the field. On the other hand, cultivar 3 produced bigger seeds and more pods than average, with larger differences between treatments and stronger variability across the field.

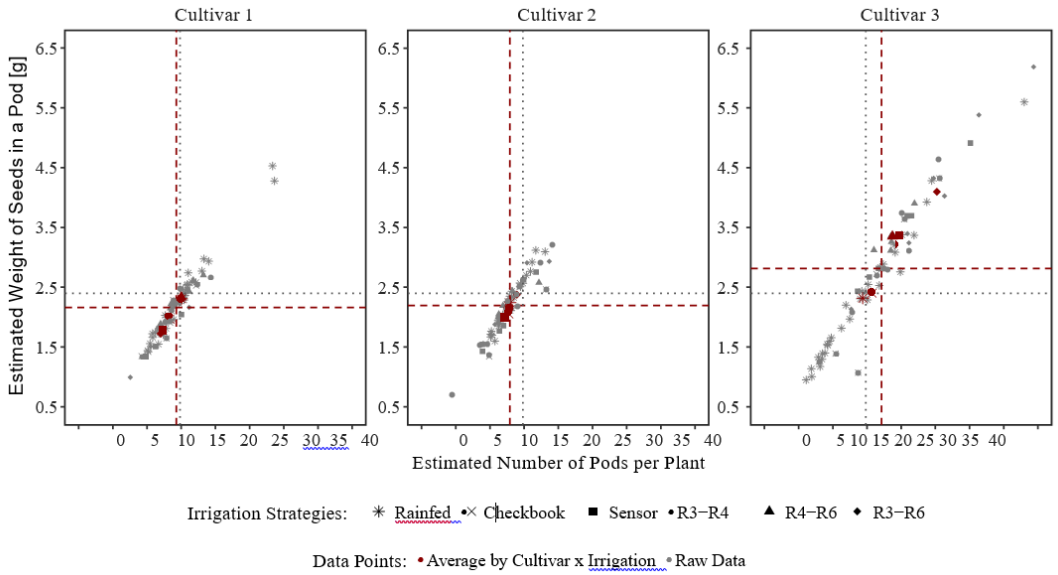


Figure 8: Estimated weight of seeds per pod as a function of the estimated number of pods per plant by cultivar. The grey points represent the original data collected along the spatial transects. The red points represent the average for each irrigation treatments, by cultivar. The grey dashed lines represent the grand averages across irrigation treatments and cultivars. The red dashed lines represent the averages across irrigation treatments for each cultivar.

Higher yields were correlated to more pods per plant and heavier seeds (Figures 9 and 10), which suggested that soybean response to excessive water stress was to produce both less pods and smaller seeds.

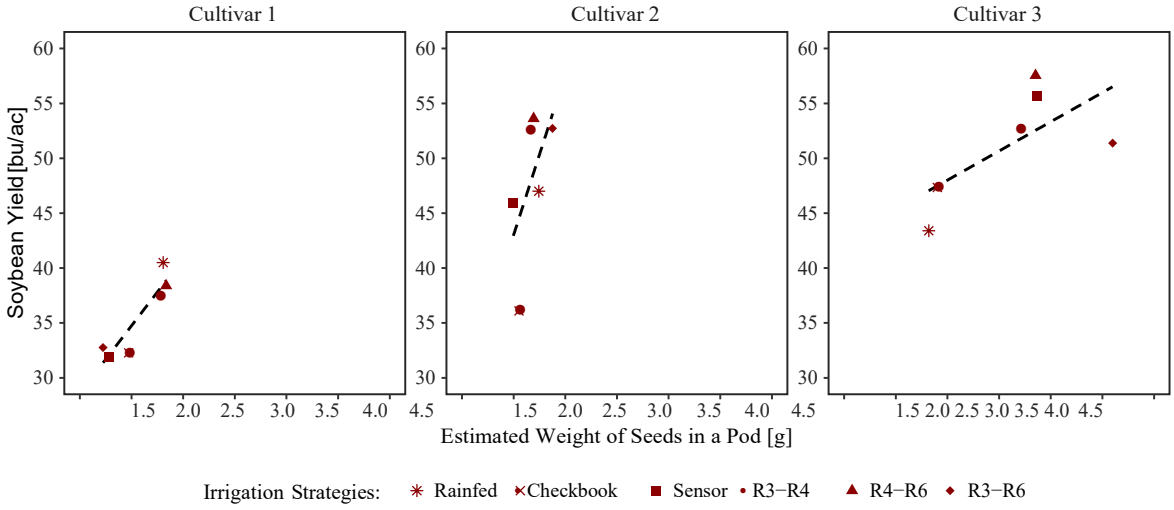


Figure 9: Relationship between soybean yield and the estimated weight of seeds per pod for each irrigation treatment, by cultivar.

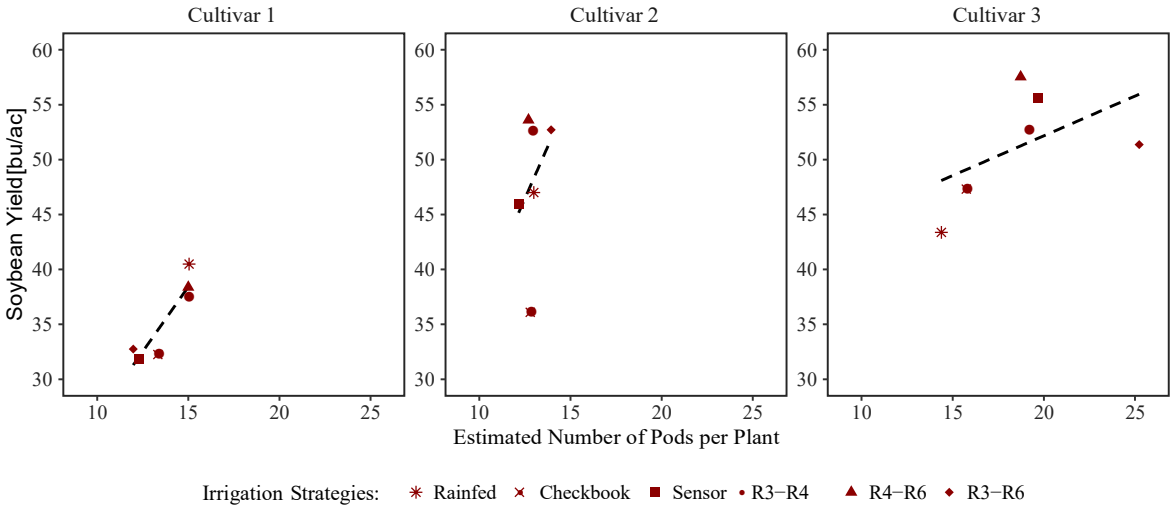


Figure 10: Relationship between soybean yield and the estimated number of pods per plant for each irrigation treatment, by cultivar.

Vegetative Development

Leaf Area Index (LAI) was measured throughout the growing season to characterize soybean vegetative development along the two spatial transects (Figure 11). LAI values at given time varied with cultivar and irrigation treatments.. Maximum LAI values were obtained between 11 and 13 weeks after planting.

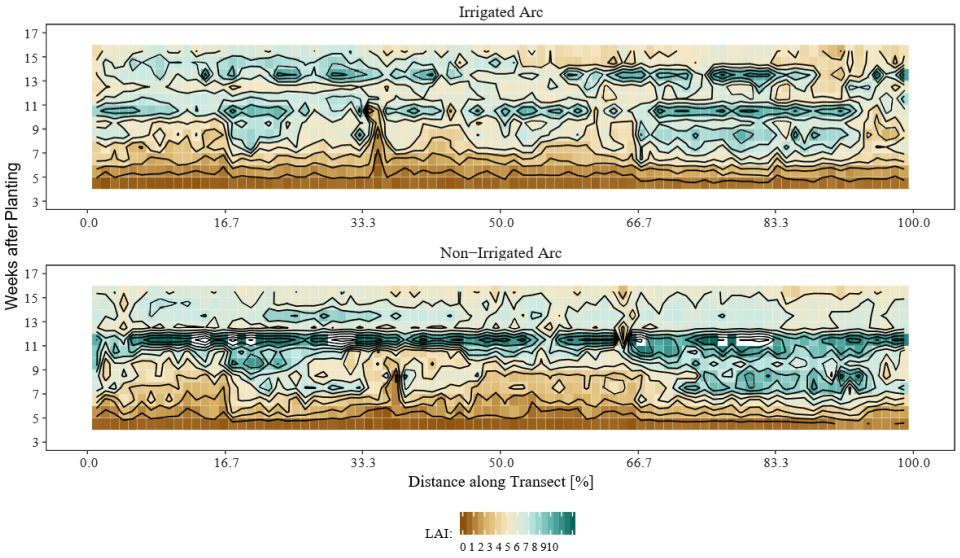


Figure 11: Evolution of Leaf Area Index along the irrigated and non-irrigated arcs throughout the growing season.

Research published in literature suggested that yield correlated with higher LAI values up to a certain point beyond which yield start to reduce with increasing LAI as excessive vegetative uses resources, which cannot be allocated to pod, and seed development. In our study, maximum LAI observed along the two spatial transects ranged from 5 to 10 across cultivars (Figure 12). No correlations were identified between yield and maximum LAI in cultivar 1. However, in cultivar 2 and 3, higher yields were correlated to lower LAI values

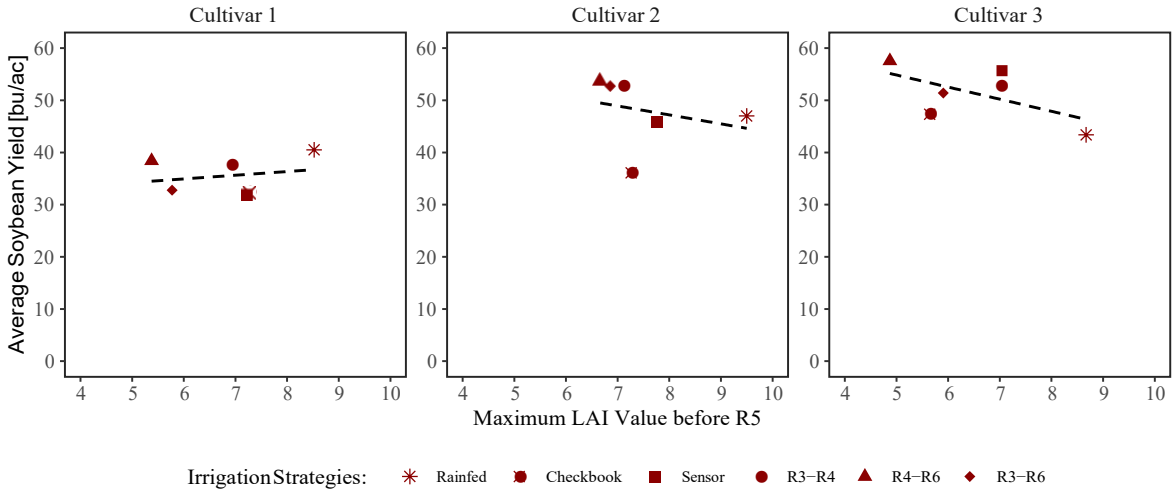


Figure 12: Yield relationship to maximum Leaf Area Index Values by cultivars

Drone Image Assessment to Improve Variable Rate Irrigation

T. Knappenberger, A. Poncet, B. Ortiz, J. Shaw, C. Brodbeck

Introduction

Drones equipped with cameras provide farmers with a relatively low-cost solution to monitor crop development at high spatial and temporal resolution. Research is being conducted across the US to integrate remote sensing data from drones to the agricultural management decision making process. Work conducted in other regions has already demonstrated the possibility of using drone imagery to improve the efficiency of current production strategies. However, minimal work has been done in Alabama.

In 2015, a field-scale, variable-rate pivot irrigation system was installed at the E.V. Smith Research and Extension Center. Cotton, corn, and soybeans are rotated on arc-shaped plots to study variable rate irrigation effects (combined with other inputs) on yield. In 2018, drones equipped with RGB and multispectral cameras were flown every 7 days during 12 consecutive weeks. Ground truth data were also collected to correlate drone data to in- field measurements at the time of flight. The objectives of this project were to measure the accuracy of multispectral drone imagery and investigate the relationships between the multispectral data, treatments, soybean yield, soil physical properties, and ground truth measurements collected at the time of flight.

Material & Methods

Drone Data Collection

Multispectral data were collected using a fixed-wing eBee Plus drone equipped with a Parrot Sequoia multispectral camera. This camera measures surface reflectance in 4 channels - green, red, red-edge, and near-infrared. Data were used to calculate to calculate four indices: the Normalized Difference Vegetation Index (NDVI, equation 1), the Green Normalized Difference Vegetation Index (GNDVI, equation 2), the Normalized Difference Red Edge Index (NDRE, equation 3), and the Chlorophyll Vegetation Index (CVI, equation 4). Higher NDVI, GNDVI, NDRE, and CVI values indicated healthier canopies. Multispectral imagery was collected on May 25, May 31, June 6, June 12, June 21, June 26, July 5, July 10, July 17, July 24, July 31, and August 7, which corresponded to the following overall soybean growth stages: V1, V2, V3, V4, V5, V6, R1, R2, R3, R3, R4, and R5. Correlations coefficients were computed to evaluate yield relationships to weekly drone data.

$$NDVI = (NIR - Red)/(NIR + Red) \quad (1)$$

$$GNDVI = (NIR - Green)/(NIR + Green) \quad (2)$$

$$NDRE = (NIR - Red\ Edge)/(NIR + Red\ Edge) \quad (3)$$

$$CVI = (NIR \times Green)/(Green^2) \quad (4)$$

Radiometric Calibration

Drones collect multiple small images along the flight path. These images must then be processed post data collection to obtain one image covering the whole study area. Horizontal and vertical accuracy of the processed image is ensured using ground control points on the edge of the field. Measurement accuracy is ensured by performing a geometric and radiometric calibration of the data.

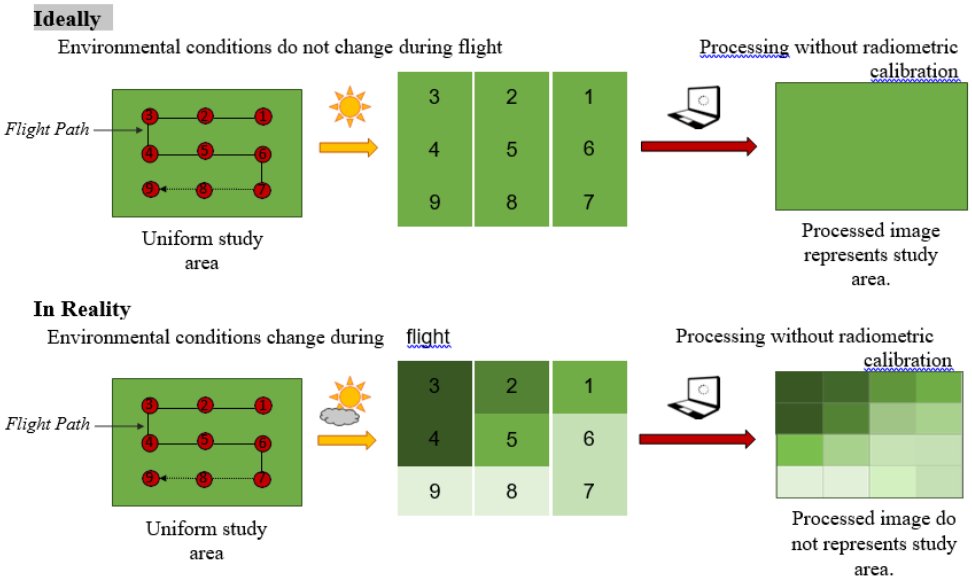


Figure 1: Example to illustrate the importance of performing a radiometric calibration to improve the accuracy of drone imagery.

The geometric calibration is usually performed by the software processing the raw images to correct for the errors associated with the geometric properties of the camera – pixel deformation on the edge of the image, changes in flight altitude and drone roll, yaw, and pitch. The radiometric calibration requires additional steps to be taken at the time of flight to correct for the errors associated with changes in environmental conditions – mainly sunlight occurring between and during flight

(Figure 1). Radiometric calibration of drone data can be performed on the small raw images prior data processing, during data processing, or after data processing. Different methods have been used in literature for the radiometric calibration of drone data, but little research has been conducted to evaluate their respective accuracy. Hence, data collected during 7 different flights were processed using five different radiometric calibration methods:

- Method 1: radiometric calibration was performed during data processing to account for changes in environmental conditions occurring between flights,
- Method 2: radiometric calibration was performed during data processing to account for changes in environmental conditions occurring both between and during flight (de- fault method for the Parrot Sequoia camera),
- Method 3: radiometric calibration was performed on the small raw images prior data processing,
- Method 4: radiometric calibration was performed using method 2 and further cali- bration was computed after data processing, and
- Method 5: radiometric calibration was performed after data processing.

Targets of known reflectance were placed across the study area to calibrate drone images using methods 3 to 5 (calibration targets). A second set of targets was also placed across the study area to evaluate measurement accuracy for all methods (validation targets, Figure 2.)

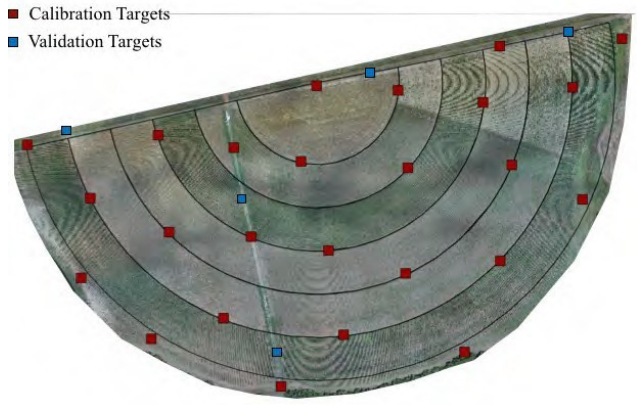


Figure 2: Location of the calibration and validation targets placed across the field in 2017 to evaluate the accuracy of selected radiometric calibration methods.

Results

Radiometric Calibration.

Statistical analysis was computed to evaluate drone image accuracy and compare the five different radiometric methods selected for this study. Results demonstrated that methods 4 and 2 both maximized drone image accuracy. Method 2 was easier to implement, and all maps and statistical analysis were computed using the drone images processed with radiometric calibration method 2.

Multispectral Vegetation Indices.

Several maps were created to show the variability of NDVI, GNDVI, NDRE, and CVI across the experimental field throughout the growing season. Results demonstrated that all veg- etation indices did not vary much across the experimental site at a given date. NDVI and GNDVI values increased during early soybean development and remained constant past R2- R3 (Figures 3 and 4). NDRE and CVI values increased during early soybean development, maximized at R3, and started to reduce with the beginning of senescence (Figure 5 and 6).

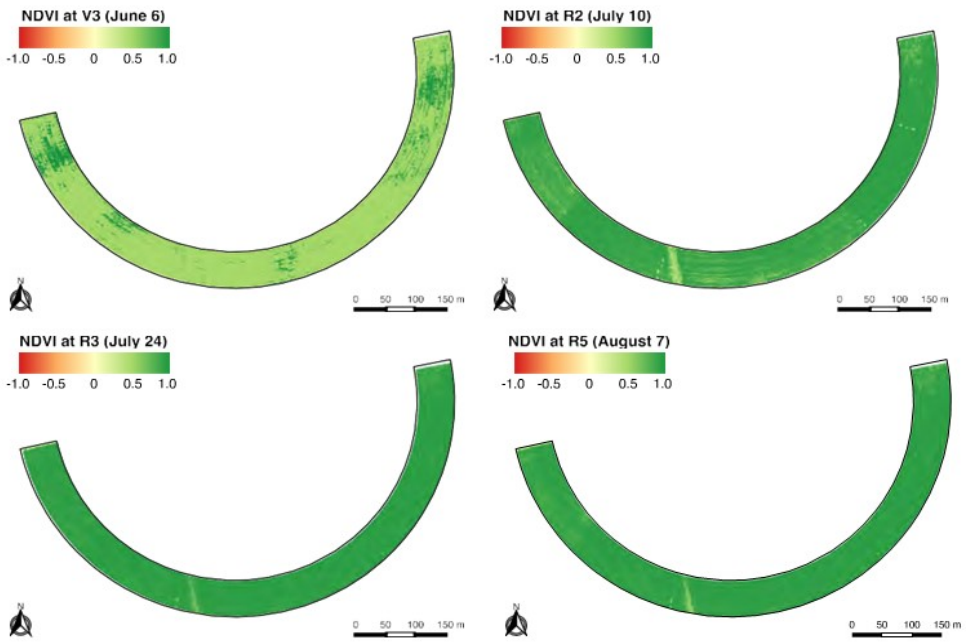


Figure 3: Example of NDVI maps computed using drone imagery to monitor soybean devel- opment in our experimental field throughout the 2018 growing season.

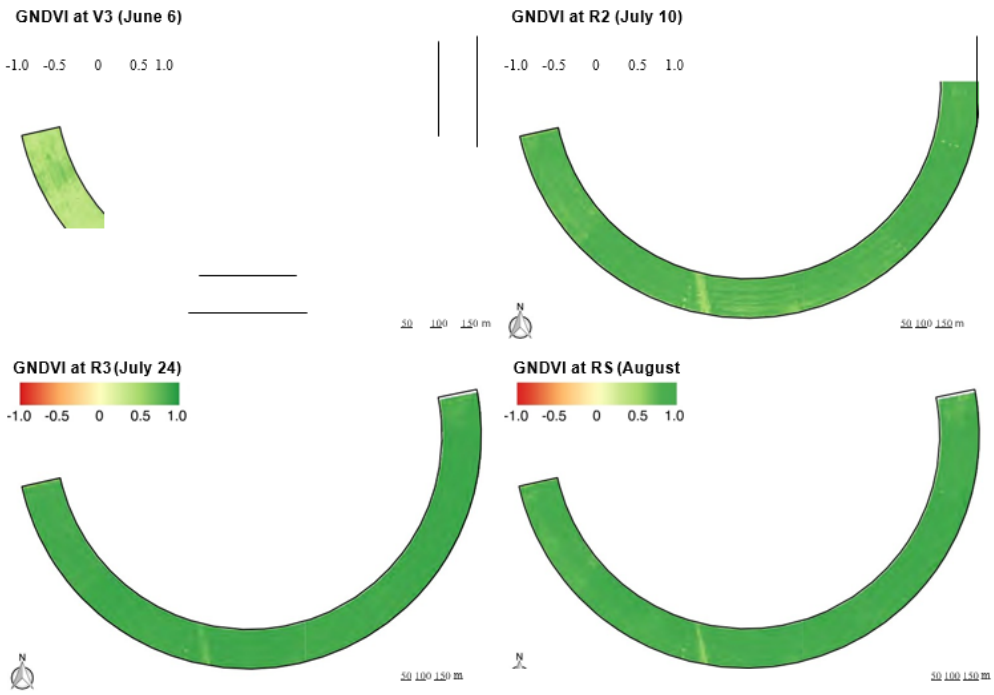


Figure 4: Example of GNDVI maps computed using drone imagery to monitor soybean development in our experimental field throughout the 2018 growing season.

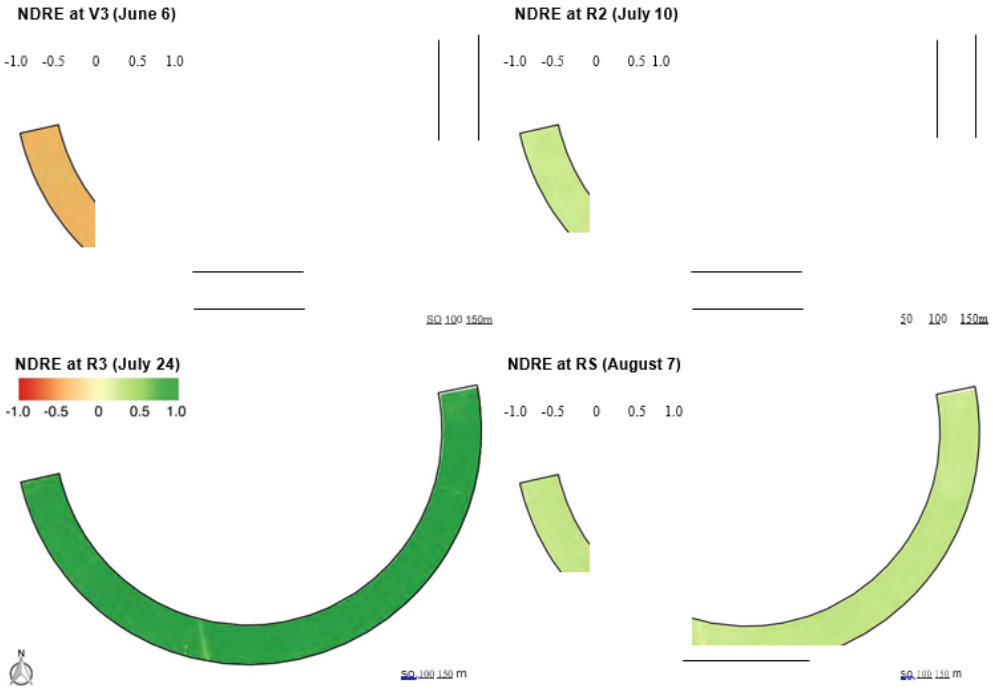


Figure 5: Example of NDRE maps computed using drone imagery to monitor soybean development in our experimental field throughout the 2018 growing season.

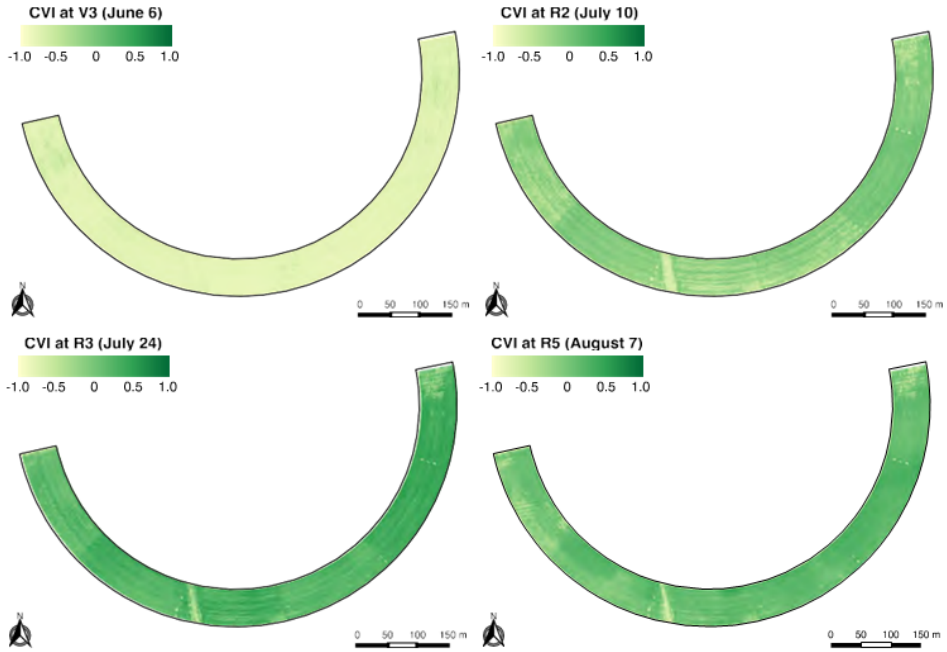


Figure 6: Example of CVI maps computed using drone imagery to monitor soybean development in our experimental field throughout the 2018 growing season.

No correlations were identified between soybean yield and the different vegetation indices throughout the vegetative stages. Yield was moderately correlated to NDVI and GNDVI from R3 to R5. Yield was moderately correlated to NDRE and CVI at R3 and R4. Strongest correlations were observed between yield and NDVI and yield and GNDVI than between yield and NDRE and yield and CVI.

Date	Growth Stage	NDVI	GNDVI	NDRE	CVI
May 25	V1	0.02	-0.03	0.13	-0.07
May 31	V2	0.03	0.10	0.25	0.13
June 6	V3	0.00	-0.04	-0.01	-0.10
June 12	V4	0.03	0.02	0.07	-0.07
June 21	V5	0.01	-0.06	0.00	-0.24
June 26	V6	0.08	0.05	-0.10	-0.12
July 5	R1	0.04	-0.09	-0.30	-0.33
July 10	R2	0.16	0.12	0.07	-0.18
July 17	R3	0.32	0.33	0.18	0.06
July 24	R3	0.28	0.34	0.28	0.31
July 31	R4	0.32	0.37	0.31	0.31
August 7	R5	0.31	0.30	0.01	0.09

Soybean Breeding- Cultivar Development

J. Koebernick

This project has helped support an active breeding program at Auburn University. It has helped increase efforts in soybean activities as well as research projects. A technician was not hired as the funding was being requested from both cotton and soybean. However, a postdoctoral candidate was identified and started the program in January 2019. A MS student started in the fall of 2018 and is investigating the defense mechanisms of target spot resistance and its relationship to iron chlorosis tolerance.

This program has not had a research focus in several years, therefore I have spent time in 2017 and in 2018, making crosses and generating material. Part of this has consisted of crossing different maturity groups, which required selections to be made in 2018 strictly on maturity. The intent is to develop similar lines that only differ by maturity. The primary objectives of the soybean program have been evolving but consist of disease resistance, improving seed quality, iron chlorosis tolerance and while maintaining yield.

Disease- Emphasis has been on target spot resistance, understanding the defense mechanism and determining if it is truly resistant. Crosses between resistant and susceptible lines were advanced this year and in 2019, molecular studies will commence. Understanding the resistance mechanism will provide insight into breeding for other foliar diseases as well.

Seed Quality- *High Oleic*- In 2017, ~800 plants were labelled and leaf samples were collected individually. Once the plants matured, each plant had the seed removed and placed into separate paper sacks. In 2018, the DNA, of the 800 plant leaves, was extracted over the summer and are being processed at UGA, with the assistance of Zenglu Li, the molecular soybean breeder. The DNA will let us know which plants have the high oleic markers and then we will go back and select the seed to be planted in 2019. *Protein/amino acid*- This year we have ventured into breeding for increased nutritional properties with primary emphasis on animal feed. New connections are in process with both AU poultry and fisheries departments in order to understand the scope/potential of these opportunities.

Iron Chlorosis tolerance- In 2018, breeding lines were identified as having moderate resistance to alkaline soils. Two of this lines were incorporated into the breeding program by crossing into existing material.

We participated in the USDA uniform and preliminary tests in 2018. Maturity groups ranged from Group IV Late, VE and VL in Belle Mina; VE, VL, VI were tested in the Uniform variety test and

IVL, V and VI in the preliminary test. Maturity groups for Fairhope were VI, VII and VIII in Fairhope. There were 12 USDA trials in total.

Other aspects of the program focus on cultivar by management strategies. The past two years the project has collaborated with Thorsten Knappenburger to determine if cultivars vary by irrigation needs. This last year we also assisted Beth Guertal on investigating phospite fertilizer.

Collaboration/Connecting: I participated in a NSF grant proposal with a researcher from the University of Alabama, using UAV remote sensing to investigate soybean roots. The project was not funded but it enabled us to make a connection and collaborate on future 2019 projects. I attended the USB board meeting in Mobile, the Soybean Breeders workshop in St Louis, the southern Soybean Pathology meeting in Pensacola and the Soybean Breeders tour in Athens, GA. These opportunities were critical in understanding the soybean industry and learning what areas of research are of interest in both the US and in Alabama. It also allowed the opportunity to engage at length with a several soybean growers, breeders and researchers from other states.

II. Fertilizer Management

Effect of Rhizobial ACC Deaminase on Soybean Root Nodulation

Y. Feng and D. Delaney

Introduction

Rhizobia-soybean symbiosis is a complex process and rhizobial entry to the soybean roots in fact causes stress to plants. Plants respond to rhizobial infection by releasing stress hormone, ethylene, which has a negative impact on nodule formation. 1-Aminocyclopropane-1-carboxylate (ACC) is the immediate precursor of ethylene in plants. Some rhizobia produce an enzyme, ACC deaminase, that can break down ACC and thereby lower ethylene concentrations in the plants. It has been reported that rhizobia with ACC deaminase activity improve nodulation by regulating ethylene levels in plants. The objective of this study was to determine if ACC deaminase activity can be used to evaluate the effectiveness of commercially available rhizobial inoculants.

Results

We obtained three soybean inoculants from commercial sources in 2018: HiStick N/T, N-DURE and N-Charge. Pure cultures of rhizobia were isolated from these inoculants using yeast extract-mannitol-Congo red agar (YMA). Slurries of inoculants were streaked on YMA plates initially. After incubation for 7 days, single colonies were selected and streaked on fresh YMA plates repeatedly until pure cultures of bacteria were obtained. One isolate was obtained from each inoculant. We have previously obtained seven isolates from other commercial inoculants (i.e., NitraStick-S, Cell-Tech, Optimize, Vault-SP, Vault-NP, Vault-LVL and RhizoStick). All 10 isolates were inoculated into a mineral medium with ACC as the sole nitrogen source in order to evaluate the presence of ACC deaminase activity. Unfortunately, none of the 10 isolates grew in this ACC-containing medium, indicating a lack of ACC deaminase activity.

We have tested a protocol for quantifying ACC deaminase activity. We also conducted a search of the genome sequence of *Bradyrhizobium japonicum* in the National Center for Biotechnology Information database and are in the process of developing a PCR-based method to detect ACC deaminase gene associated with *Bradyrhizobium japonicum.*

Exploring Phosphite Fertilizer for Soybean: Fungicide or Fertilizer?

B. Guertal, J. Koebernick, and A. Gamble

A. Research Idea:

Phosphite is a generic name used to describe alkali metal salts of phosphorus acid (H_3PO_3). The most common phosphite in fertilizer or fungicide is potassium phosphite. Phosphite is NOT phosphate, and only phosphate is included in fertilizer labels as a fertilizer nutrient (in the US). However, there are many fluid products that also contain phosphite, and they are often marketed with claims of 'stress reduction' properties, since direct fungicidal claims cannot be made without EPA labeling.

Phosphite is converted to phosphate in the soil, but little work has been done to quantify how quickly this happens. Our work at Auburn University has shown a ½ life for phosphite to phosphate conversion (in a loamy sand soil) of 1 to 2 months. But – the application of phosphite has issues. Phosphite may be toxic to crops in the first cropping cycle. Many reading this proposal may remember extensive crop losses in the 1970s from when phosphite was incorrectly applied. Regardless, phosphite is being applied, although typically at low rates, for a perceived fungicidal effect, and there may also be some subsequent fertilizer effects.

So, what was the justification for this proposal? First, we wanted to explore if foliar applications of phosphite could increase soybean yield, possibly through fungicidal benefits. Second, we wanted to see if this phosphite eventually also provides a fertilizer P boost, as it is converted into phosphate, most likely by a second year of cropping. We need to do this over a range of soil P, because when soil P is low the phosphite may be harmful to the crop. However, we do not know (nor does any literature show) the typical soil-test P levels and phosphite application rates at which such damage may occur. Our preliminary field work indicated that soil-test P can become very low, and all we ever saw was the positive effects of the applied fluid phosphite acting as a fungicide. Because there are many commercial fluid phosphite products in the fertilizer market, the objective of this research was to: 1) examine commercial phosphite fertilizers for their effect on soybean, and, 2) determine if recommended rates of commercial phosphite products affected soybean growth across a range of soil P content.

B. Objectives

- Determine how the application of fluid phosphite fertilizer affected soybean growth, when applied in both soil and foliar-applied forms, and determine if any reductions in disease are found.
- Conduct Objective 1 over a range of soil-test P (extractable orthophosphate) levels.

Note: this report covers the field testing, as greenhouse work has not yet started. That greenhouse work will evaluate various phosphite materials at various rates.

C. Methods and Materials

The experiment will have two phases: a greenhouse phase and a field study. This report is for the field study.

The field study was conducted at the E.V. Smith Research Center located in Tallassee, AL. Soybean (Credenz CZ5242LL) was planted in a clean-till system on June 5th 2018. Plots consisted of 4 rows (36 inch spacing) with a plot length of 25 feet. There were 4 replications of each treatment. Background soil testing indicated a soil pH of 6.0, and Mehlich extractable P of 31 lb/A (a Medium test) and K of 110 lb/A (a High test). Recommended P_2O_5 was 40 pounds per acre, with no K or lime recommended. The plots were harvested on October 18th, 2018.

Treatments were a factorial combination of soil P (applied as triple superphosphate) at rates of 0, 20, 40 and 80 pounds P_2O_5 per acre (this bracketed the recommended rate). These were combined with application of phosphite (using the Harrells brand TitlePhyte, which was 38% P). There were three applications of the phosphite material, applied on August 6th, August 20th and September 4th to supply 1.5 lb P/A as phosphite P. Immediately before each application the height of 3 randomly selected plants was taken. At 48 hours after application phytoxicity measurements were taken (1-5 scale, visual) and most recently emerged whole leaves were collected for subsequent P analyses (still being completed). There was never any sign of phytoxicity or damage after spraying the phosphite, and so none of that data is included in this report. Additionally, plant height was rarely affected by either phosphate or phosphite.

Results:

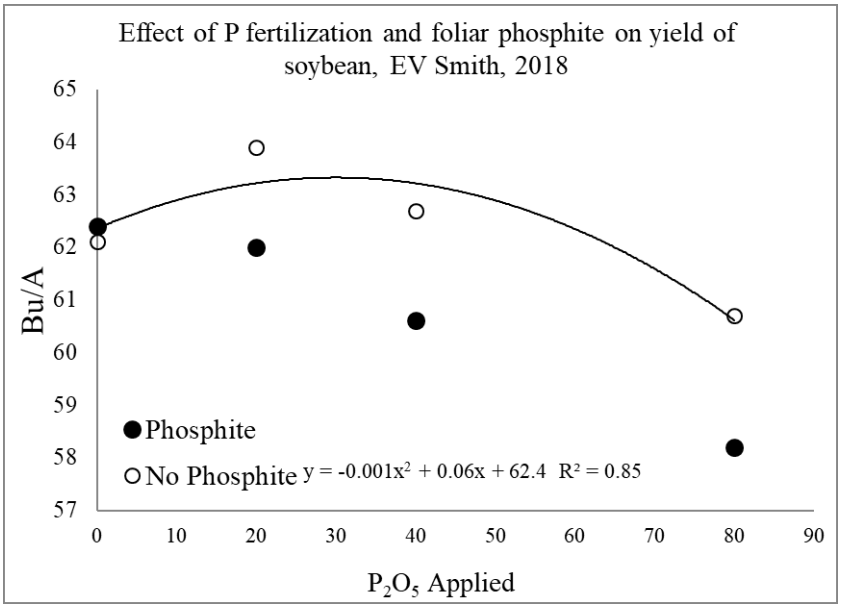


Figure 1. Yield of soybean as affected by rate of P fertilization (as triple superphosphate at planting) and foliar phosphite.

Comments:

- Yield of soybean was slightly increased when 20 pounds of P_2O_5 was added. This was in line with soil-test recommendations, which recommended 40 pounds. Yield was not improved by the addition of any addition phosphate fertilizer.
- Yield was decreased when phosphite was applied, and yield reductions increased as the rate of P fertilization increased.
- We saw no benefits from the addition of phosphite to the soybean production system.

Next Steps:

- Plant a second year of the work, to determine if similar results occur.
- Complete the greenhouse work to better determine soil-test P at which such negative effects might occur, due to the application of phosphite.

Funding Request:

We are not requesting additional funding. We will finish the greenhouse work with the current funds, and conduct the second year of research for our own interest. At this point we cannot recommend phosphite application to soybean for any possible fungicidal benefits, and do not seek additional monies.

Benefits of Residual Fertilizer on Soybeans Following Two Years of Corn Production Using Poultry Litter

M. Hall, T. Reeds, and T. Sandlin

Yield response of soybeans to the residual fertility following the year after corn received different fertility treatments. There was no significant difference in yield with any treatment.

2017 Corn Fertility Treatment per acre	2018 Soybean Yield per acre
2 tons of poultry litter	68.70
Fertilized as recommended by Auburn soil test	68.34
Commercial fertilizer equal to 2 tons poultry litter	68.68

Dry Land

2017 Corn Fertility Treatment per acre	2018 Soybean Yield per acre
2 tons of poultry litter	68.20
Fertilized as recommended by Auburn soil test	69.18
Commercial fertilizer equal to 2 tons poultry litter	67.00

This test was conducted at the Tennessee Valley Research and Extension Center using a randomized complete block design and 4 replications per treatment.

2018 Soybean Systematic Optimization of Yield –Enhancing Applications (SOYA)

E. McGriff, D. Delaney, T. Sandlin, and H. Farms

Test Location: Limestone County

Planted: May 3, 2018 Harvest Date: October 22, 2018

Tillage: Conventional Seeding Rate: 130,000

Previous Crop: Corn Soil Type: Decatur Silty Loam

Hybrid: USG 74A74

Plots were randomly replicated four times with four 30-inch rows. Row length was 30 feet. Two inside rows were harvested with a plot combine and weighed. Tissue samples were taken bi-weekly. Yield was adjusted to 13.0 % moisture. Two tons of chicken litter per acre was applied in the fall. Herbicides applied post-emerge were glyphosate followed by a glyphosate + Engenia (dicamba). Stratego YLD + Dimilin + a pyrethroid were applied at R2.

Table 1. Limestone County, Alabama

Irrigated Soybean Systematic Optimization of Yield-Enhancing Applications (SOYA)

Treatment	Yield	Profit*
Lbs/A	Bu/A	
Foliar N applied at R2 (9.78 lbs of 46% urea; a total of 4.5 lbs of N)	97.88	+$44.34
30 lbs N applied AP as ammonium nitrate	95.83	+$15.67
30 lbs N applied R3 as ammonium nitrate	95.30	+$10.27
10 gallons 3-18-18 applied AP as sidedress (dribbled 4 inches beside the row)	94.90	-$18.17
Foliar KNO_3 applied at R2 (10 lbs of 13-0-44)	94.78	+$8.30
20 gallons 3-18-18 applied AP as sidedress (dribbled 4 inches beside the row)	94.72	-$62.50
2 tons chicken litter (AP)	94.37	-$36.07
120 lbs 0-0-60 applied at R3 (72 lbs of K)	93.63	-$20.08
120 lbs 0-0-60 applied at AP (72 lbs of K)	93.41	-$22.32
Untreated check, Auburn soil and private labs recommended no fertilizer due to high nutrient levels	92.51	0
200 lbs of KMag + 30 lbs of N (ammonium nitrate) applied at R3	92.22	-$65.28
200 lbs of KMag applied at R3	91.32	-$63.31
30 lbs of N applied at R3 as ammonium sulfate[#]	87.22	-$82.67
120 lbs of 0-0-60 + 30 lbs N (ammonium sulfate) applied R3[#]	86.26	-$116.93
Plot Average	93.11	

*Profit was derived by subtracting the cost of materials and application cost from the increase in yield (bushels per acre) times the price of which the grower sold the soybeans ($10.18 per bushel). Input costs are on following page.

[#] Significant foliar burn from ammonium sulfate may have contributed to lower yields.

Cost of two tons per acre of chicken litter spread was $55; 30 lbs of N per acre spread as ammonium nitrate was $18.13; 3-18-18 was $4.25 per gallon; 120 lbs per acre of 0-0-60 was $24.48 + $7 per acre spreading cost; 200 lbs per acre of KMag was $44.20 + $7 per acre spreading cost; ammonium sulfate was $12 per 50 pound bag + $7 per acre spreading cost; urea was $17 per 50 pound bag + $7 per acre application cost; potassium nitrate was $39 per 50 pound bag + $7 per acre application cost .

2018 Soybean Systematic Optimization of Yield-Enhancing Applications (SOYA)-Late Season

E. McGriff, D. Delaney, T. Sandlin, and T. Farms

Test Location: Calhoun County

Planted: June 15, 2018 Harvest Date: November 30, 2018

Tillage: No-Till Seeding Rate: 140,000

Previous Crop: Wheat Environment: Center Pivot Irrigation

Hybrid: Pioneer 48T27X

Plots were randomly replicated four times with four 30-inch rows. Row length was 30 feet. Two inside rows were harvested with a plot combine and weighed. Yield was adjusted to 13.0% moisture. Grower applied 350 pounds per acre of 5-20-20 before wheat crop for both wheat and soybean crops. Herbicides applied were two applications of glyphosate postemergence.

Table 1.Calhoun County, Alabama

Irrigated Late Season (soybeans behind wheat) Soybean Systematic Optimization of Yield-Enhancing Applications (SOYA)

Treatment	Yield	Profit*
Lbs/A	Bu/A	
Untreated check and Auburn Lab recommendations the same (no fertilizer recommended)	69.01	base
30 lbs N applied (AP) as ammonium nitrate	66.42	-$47.49
200 lbs of KMag applied at R2	65.84	-$78.86
120 lbs 0-0-60 (AP)	65.33	-$63.68
2 tons chicken litter (AP)	64.24	-$101.74
Private Lab Recommendations 20-40-80 applied (AP)	62.81	-$111.06
200 lbs of KMag + 30 lbs of N (ammonium nitrate) applied at R3	61.72	-$133.12
30 lbs N applied (R2) as ammonium nitrate	60.74	-$97.49
120 lbs 0-0-60 applied at R2	59.03	-$118.81
Plot Average	63.90	

*Profit was derived by subtracting the cost of materials and application cost from the increase in yield (bushels per acre) times the price of which the grower sold the soybeans ($8.75 per bushel) over the untreated check. Input costs are two tons chicken litter spread ($60);18- 46-0 ($520 per ton); 0-0-60 ($408 per ton); 33.5-0-0 ($405 per ton); KMag ($442 per ton); and $7 spreading cost.

Amount and Timing of Nitrogen Release from Poultry Litter in Soybean Production System

R. Prasad, T. Reed, and W. Birdsong

Project Overview and objectives:

Most soybean growers value chicken litter as an important slow release source of nutrient. Growers typically use 60-60-40 (lb/ton) as the total nutrients (N-P2O5-K2O) and 40-40-30 (lb/ton) as the available nutrients during first season of litter application. However, growers are poorly informed as to when and how much nutrient, primarily N, is available to plants. Through this project, we tried to answer following questions:

1. How much and when is nitrogen released after application of poultry litter?
2. Is there a benefit of applying chicken litter for soybean production? If yes, are there yield differences among different rates of application?
3. Is there an effect of chicken litter application on nutritional composition of soybean seeds?

Methods:

Field plots (four rows of 20 ft length) were established at E.V. Smith research center in Compass loamy sand. There were four treatments (Table 1) arranged in a randomized complete block design and replicated four times. Chicken litter was applied at 1, 2.5 and 5 ton/acre on 5th July (7 days prior to planting, Figure 1). Soybean cultivar AG74X8 (**maturity group 7**) was planted on 12th July, 2018 at a population of 10 seeds/ft in 36inch row spacing in **dryland condition**. The plots were maintained weed and disease free throughout the season. The crop was harvested on 29th Nov with a plot combine, percent moisture determined and plot weights converted to bu/A yield at 13% moisture. The seeds were dried and analyzed for nutrients (P, K, Ca, Mg) by acid digestion followed by ICP analysis.

Soil samples were collected from each treatment plots to estimate nitrogen release (nitrogen mineralization) rate over time. Soil cores were collected at 0-15cm and 15-30 cm depth at day 0, 1, 5, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120 and nitrate-nitrogen and ammonium-nitrogen were determined. Nitrogen mineralization was estimated by subtracting the N released in control plot from treatment plots (e.g T1-T4).

Table1: Treatments

	Treatments
T1	1 ton/acre (Chicken litter)
T2	2.5 ton/acre (Chicken litter)
T3	5 ton/acre (Chicken litter)
T4	Soybean only (control; no litter)

Figure 1 Differences in chicken litter application rate

Project preliminary results

1. How much and when is nitrogen released after application of poultry litter?

We are still working on this section. The laboratory analysis is taking time. Once we have the results we will share it with everyone. In addition, this is a work-in-progress. We will need a second year data to make a robust estimation of the N release rates and timing from application of chicken litter.

2. Is there a benefit of applying chicken litter for soybean production? If yes, are there yield differences among different rates of application?

Yes, soybean responded to chicken litter application (Figure 2). There was a 10 bu yield difference between control plots and treatment plots (Figure 3). Application rate of 5 ton/acre gave the greatest yield, however, there was no significant difference between application rates.

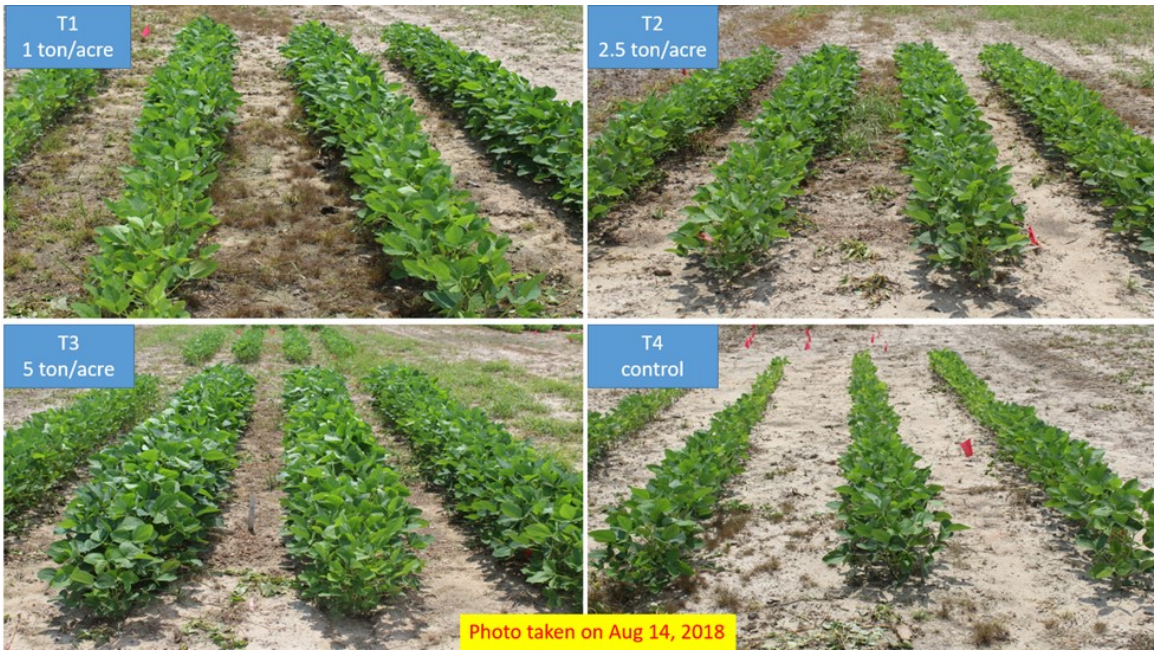


Figure 2. Visual difference in soybean growth rates between different litter application rates.

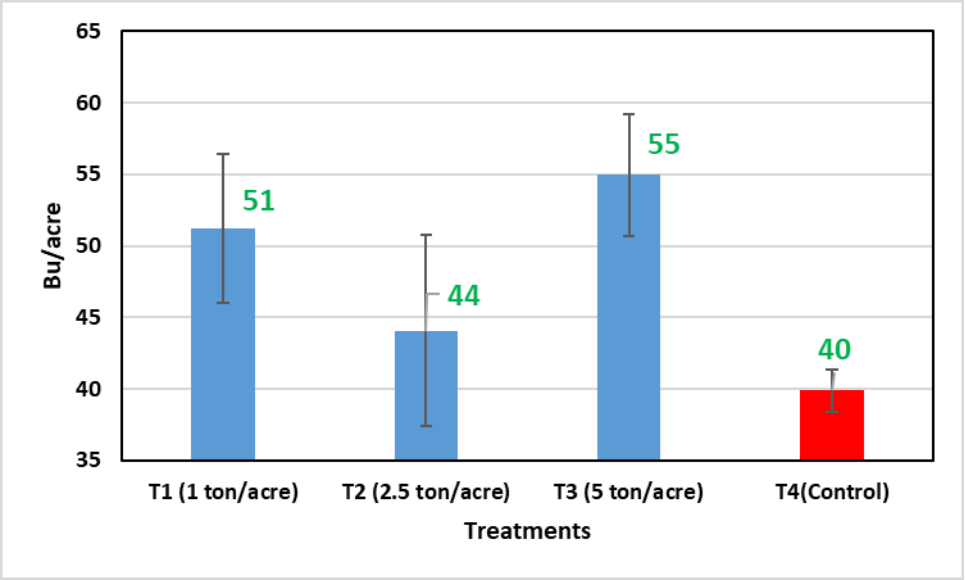


Figure 3: Soybean yield response to chicken litter application rates

3. Is there an effect of chicken litter application on nutritional composition of soybean seeds? The seed nutrient concentrations of N, P, K, Ca, Mg, S, Fe, Mn and Cu did not change between treatments except for B (Table 2). **Boron concentrations in seeds increased with increase in chicken application rate.** An important take home message from this study is that greater yield was achieved with application of chicken litter but their nutrient contents were no compromised. Nutrient concentrations in control plots were same as the chicken litter plots.

Table 2: Nutrient concentrations in soybean seed. None of the concentrations were significantly different expect for Boron

Trt	N%	P%	K%	Mg%	Ca %	S%	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)
T1 (1 ton/acre)	7.0	0.6	1.8	0.3	0.4	0.4	31.8ab	50.1	58.7	91.4	15.1
T2 (2.5 ton/acre)	6.9	0.6	1.9	0.3	0.4	0.4	33.7ab	49.9	68.7	89.8	17.2
T3 (5 ton/acre)	6.8	0.7	2.0	0.3	0.4	0.4	37.2a	46.0	64.7	97.6	16.5
T4 (control)	6.9	0.6	1.9	0.3	0.5	0.4	30.4b	50.0	57.8	88.5	15.7

A Decision Support Tool for Phosphorus Application in Soybean Fields that have a "High" Soil Test Phosphorus Rating

R. Prasad, A. Gamble, D. Delaney, and K. Stanford

Background

This project was initiated with a goal to understand the phosphorus storage capacity (SPSC) of soils (in soybean fields) that receive poultry litter or soils that have a "high" soil test phosphorus rating. The data collected from this project will help the state of Alabama modify its P index and the stringent changes proposed under code 590 of the Natural Resources Conservation Service (NRCS).

The project required the participation of Alabama farmers to voluntarily allow taking soil samples from their fields. Several promotions /campaigns (promo card (Figure1), Facebook, announcements at ALFA expo etc.) were launched to encourage farmers to participate in the program. As a courtesy, we proposed to offer free soil test reports to the farmers. Additionally, we promised to keep the names and locations of the farms confidential.

Method

Soil samples were collected at the volunteer farms (Figure 2). The soil samples were collected at several locations (4 to 7) and four depths (0-2, 2-6, 6-12, 12-24 inch) per farm, depending on the ability of the soil probe to cut through greater depths (Figure 2 and 3). The soil samples were dried, ground and extracted using extractants namely, Mehlich1, Mehlich-3, Oxalate and water, and P concentrations were determined. The relationships are currently being studied. Preliminary result is presented below.

Preliminary results

We are still working on the data analysis in laboratory. For this report we are presenting data from two farms. The data below is preliminary and used for reporting purpose only. Drawing strong conclusion is not recommended at this time.

To maintain the confidentiality of the farms, we have named the farms as Farm A and Farm B. The soils at farm A is Nauvoo and Sipsey soils whereas soil at Farm B is Orangeburg loamy sand. Soil phosphorus storage capacity was calculated for the two farms using the methods described above. As presented in Figure 3, the soil in 0-2 inch depth has negative SPSC value. When the SPSC value is negative, the soil has no more capacity to fix any additional phosphorus and the phosphorus holding capacity is exhausted. On the other hand, when SPSC value is positive, the soil has the remaining capacity to absorb/fix more phosphorus. The preliminary results indicate that:

1) The magnitude of SPSC is different between farms and soil depths. Farm B has greater negative SPSC value than Farm A.

2) The SPSC values becomes positive as we go down the soil profile. This indicates that soils at lower depths are still holding the P and preventing it from leaching to the groundwater.

Based on these two data sets, it is obvious that soils in these two farms have the capacity to hold more phosphorus. If litter is injected in subsurface soils, there should be no risk of P loss. Also, these findings will play an important role in modifying the changes proposed in 590 standards.

More participation of farmers is required to get a robust data set representative of Alabama soils. Due to wet condition in Fall, we could not collect samples as anticipated. We will continue soil sampling in Spring 2019.

Figure 1. Sample promo card used to encourage farmers to allow soil

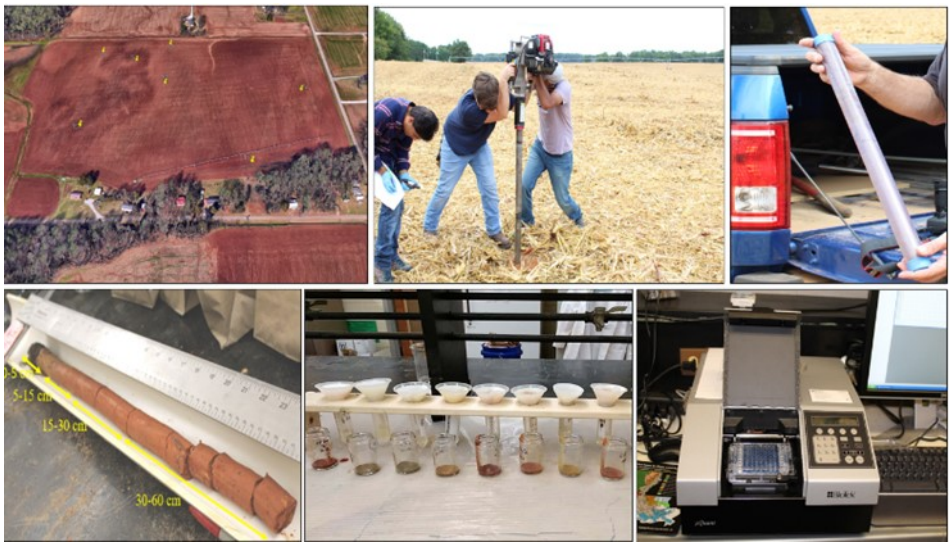


Figure 2. Soil collection and laboratory analysis of phosphorus

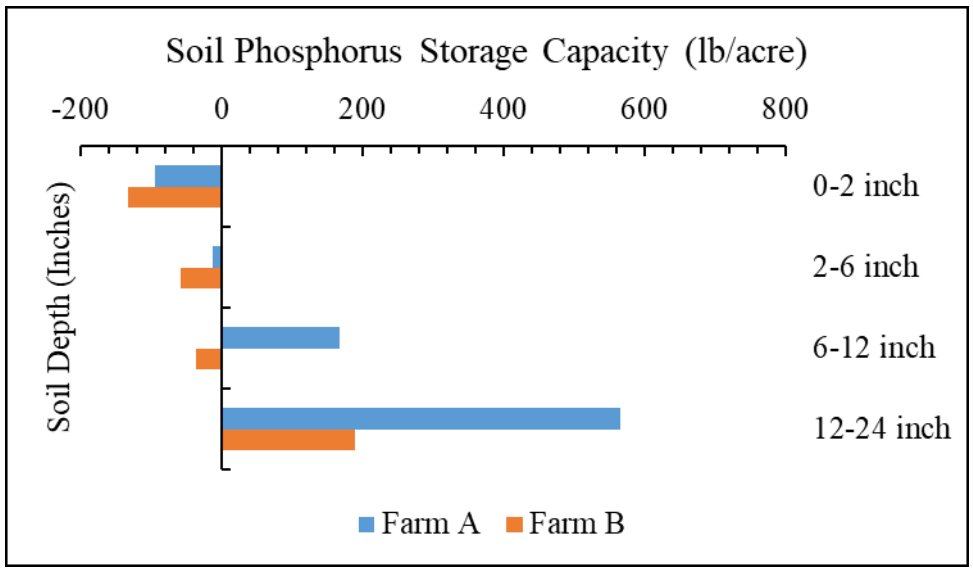


Figure 3. Comparison of soil phosphorus storage capacity of two farms.

III. Weed Management

Evaluation of Herbicide-Resistant Annual Ryegrass (*Lolium multiflorum)*

Title-Soybean: Evaluation of Herbicide-Resistant Annual Ryegrass (*Lolium multiflorum)* Non-Controlled in Burndown Applications Prior to Soybean Planting

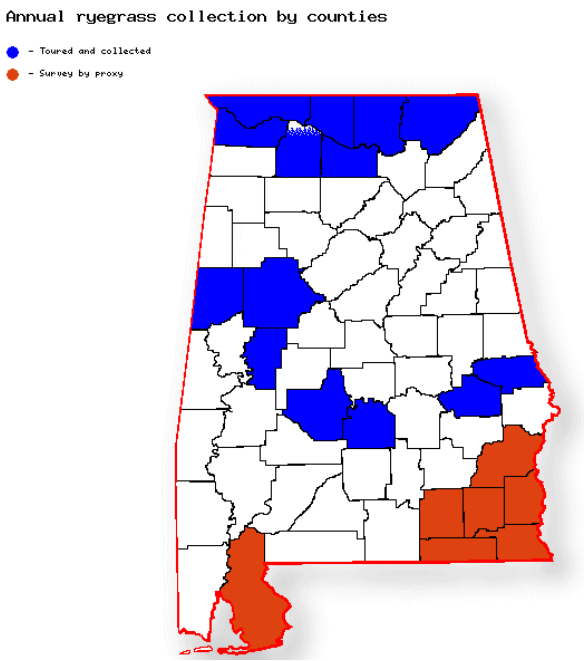
Title-Wheat and Feed Grains: Evaluation of Possible ACCase and ALS-Resistant Annual Ryegrass (*Lolium multiflorum)* in Alabama Wheat Fields

S. McElroy and T. Sandlin

Justification

Surrounding states, such as Mississippi and Tennessee, have reported extensive distribution of herbicide resistant annual ryegrass (*Lolium multiflorum*). The existence and distribution of herbicide resistant ryegrass is largely unknown within Alabama.

Methods



A survey of fields was conducted in March through May 2018 to attempt to locate annual ryegrass populations not controlled by herbicide applications (see map inset). In-person field evaluations was conducted by the investigator and collaborator throughout North Alabama and parts of central east and west Alabama by the investigator. Extension specialist and agents, as well as chemical and sales distributors throughout the southern part of the state were contacted to aid in locating non-controlled populations. Counties surveyed in person by investigator and/or collaborator were: Colbert, Dallas, Hale, Jackson, Lauderdale, Lawrence, Lee, Limestone, Lowndes, Macon, Madison, Morgan, Pickens, and Tuscaloosa. In-person surveys were primarily by word-of-mouth communication to the investigator or collaborator or driving field surveys using aerial maps to locate pasture and agronomic crop production areas. Areas survey by proxy via extension personnel or sales distributors were: Barbour, Baldwin, Coffee,

Dale, Geneva, Henry, and Houston. These survey methods were meant to "cast a wide net" in hopes capture any uncontrolled annual ryegrass population that may exist.

Results

It is the opinion of the investigator and collaborator that changes in herbicide use has likely confounded our resistance collection efforts. Herbicide usage in wheat has shifted to Axial (pinoxaden) and Powerflex (pyroxsulam), which is likely controlling any Hoelon (diclofop)-resistant annual ryegrass that may exist. It was therefore difficult to find annual ryegrass in wheat fields surveyed. In addition, burndown applications prior to planting summer crops now utilize a protoporphyrinogen oxidase (PPO)-inhibiting herbicide (such as Sharpen - saflufenacil) or photosystem I inhibitors (paraquat) in addition to glyphosate. Additional herbicides are likely masking annual ryegrass resistance to glyphosate and increasing the difficulty in locating resistant populations. Despite these restrictions, a total of 37 populations were able to be collected from the survey areas. Plants were only collected from the areas toured and collected by the investigator and collaborator. No plants were collected from areas surveyed by proxy.

Of the 37 populations collected, all were propagated for seed in the Auburn University Weed Science Greenhouse. Seed were collected May and June 2018. Seed were placed in cold storage (4 C) until September. Seed were planted in September and glyphosate at 0.5 lb ae/ a (equivalent to 1 pt/a RoundUp products containing 4 lbs ae/gal) or Hoelon at 2 pts/a were applied to 10 plants from each population collected approximately 6 weeks after germination.

Of the 37 populations collected, only three displayed signs of possible resistance. Populations were 'Ripley' (collected along Ripley Rd. in Limestone County), 'Blackburn' (collected at the corner of Blackburn Rd. and Hwy 72 in Limestone County), 'Hyundai' (collected along Hwy 31 adjacent to the Hyundai Automotive Plant in Montogomery County). Ripley was collected from a fallow field with obvious burndown application, Blackburn was collected from a wheat field, and Hyundai was collected from a roadside with known use of glyphosate. Blackburn was Hoelon-resistant, Ripley was resistant to both Hoelon and glyphosate, and Hyundai was resistant to glyphosate. No other populations collected were resistant to Hoelon or glyphosate.

Observations

While only observational, the investigator and collaborator noted that there was considerably more annual ryegrass in both wheat and fallow fields in northwest Alabama than in any other part of the state. Further, it was our opinion that the amount of annual ryegrass increased moving west from I-65 both north and south of the Tennessee River in Colbert, Lawrence, Limestone, and Lauderdale counties. A follow up field survey was conducted in early January 2019 of these areas and a similar pattern was observed as in 2018.

In-person surveys of Baldwin County were conducted by the investigator in 2016 prior to development of this grant. Due to perceived changes in herbicide usage it was difficult to find annual ryegrass in wheat fields or fallow fields after burndown applications. Twelve populations were collected from Baldwin County primarily along county roads 104 and 49 in wheat, fallow, and roadsides. Two populations collected from roadsides were found to have elevated tolerance to glyphosate.

Conclusions: *Both Hoelon and glyphosate resistant annual ryegrass are present in Alabama.* Such resistance could one day lead to an expansion of uncontrolled annual ryegrass both in small-grains and summer agronomic crop production. However, based on in-person surveys of select regions, communications with extension personnel, chemical sales distributors, and growers, *the problem is seen as minor and no further request for funding for this project will be made at this time.* It is our opinion that the majority of the problem is concentrated in the Northwest part of the state with possible movement occurring from Mississippi and Tennessee into Alabama. Lastly, herbicide-resistant annual ryegrass, primarily to glyphosate, is present is Alabama roadsides and could likely serve as a means of expansion and spread of the problem in the future.

Future Research

Blackburn, Ripley, and Hyundai populations are currently being propagated for a rate titration screen to be conducted later in spring 2019 to determine the exact degree of resistance to herbicides. Further, DNA sequencing will be conducted to determine if known mutations are conferring the observed resistance.

More samples are wanted

The investigator and others at Auburn University remained committed to surveying the entire state for not only herbicide-resistant annual ryegrass but any suspected herbicide resistant weeds. Anyone can contact Dr. McElroy at mcelroy@auburn.edu or cell 334-740-9781. Dr. McElroy or

his staff will come to collect any and all weeds that may be herbicide resistant. Rapid detection is key to containing herbicide resistance. We are open and actively seek your communication to us.

Evaluation of Palmer Amaranth Control with PPO Herbicides Before and After Dicamba Application

T. Sandlin

Location: TN Valley Research and Ext. Center (TVREC)

Application Volume: 15 GPA

Justification and Methods

In 2016 PPO resistant Palmer amaranth was confirmed in Alabama. Numerous phone calls from growers were received by extension personnel. Samples from several suspect fields were sent off for testing. In 2017, few if any phone calls were received regarding these issues. This was due to Auxin herbicides being labeled for use in crop, and excellent weed control being achieved. Two applications of dicamba in soybeans were common in 2017. Multiple applications of the same mode of action intensifies selection pressure and can lead to resistance. Data generated at other universities has indicated that an application of a PPO herbicide following an application of dicamba can result in control Palmer amaranth. Data has also indicated that when an application of a PPO herbicide is made following an application of dicamba on PPO resistant Palmer amaranth, that some of the resistance mechanism is reversed, allowing the plant to be susceptible the herbicide. We would like to test these situations in Alabama.

Tests were conducted in a location set aside for weed science research TVREC because no PPO resistant Palmer amaranth could be located in farmer's fields. Palmer amaranth pressure was severe. Plots were established to determine if "oversized pigweed" (8" and larger) could be controlled with treatments listed in table 1.

Results and Discussion

Large Palmer amaranth were initially sprayed with the treatments in table 1. with and application volume of 15 GPA. After visually rating plots at 14 days after application, it was very apparent that minimal control was achieved for all treatments. This was due in part because of three factors (1) Palmer pigweed were too large (2) sprayer pressure needed to be greater-spray pattern was inconsistent and minimal coverage was achieved (3) The weed canopy was so dense that all weeds were not contacted by spray droplets. Due to these factors, we waited for a second flush of new weeds and repeated the study and increased sprayer pressure. Large Palmer amaranth (8" and larger) that had initially been sprayed were rated as well as smaller Palmer amaranth (6" and less) that emerged in the new flush of weeds. Ratings for this test are listed in table 1.

Table 1. Large and Small Palmer amaranth percent control 14 and 28 days after application

Treatments	Large Palmer % Control 14 DAA	Large Palmer % Control 28 DAA	Small Palmer % Control 14 DAA	Small Palmer % Control 28 DAA
22oz Xtendimax FB 22oz Xtendimax @ 7 Days	45	65	90	90
22oz Xtendimax FB 16 oz Reflex @ 7 Days	42	65	90	90
16oz Reflex FB 22oz Xtendimax @ 7Days	37	60	90	90

Summary

- Some large Palmer amaranth was controlled but not at an acceptable level. These results along with data from other universities confirm that there are size limitations with respect to dicamba applications and follow up treatments for Palmer amaranth control.
- Much better control was achieved with the same treatments for small Palmer amaranth.
- Xtendimax followed by Reflex seven days later resulted in equal control to Xtendimax followed by Xtendimax.
- This allows for multiple modes of action to be used and helps alleviate selection pressure for Xtendimax and similar products and helps to reduce resistance to these products.

Soybean Varietal Response to Suflufenacil (Sharpen Herbicide)

T. Sandlin and D. Delaney

Location: TN Valley Research & Ext. Center (TVREC)

Justification and Methods

Knowing that varietal sensitivity does exist, we have found that an in-field screening of commonly planted soybean varieties for this area is a valuable tool. Thirty-eight soybean varieties were successfully screened for tolerance to saflufenacil in 2018. Please note that these results are based on silt loam soils. These plots were sprayed one day after planting with 1.0 oz, 1.5 oz, and 3.0 oz/Acre of Sharpen respectively, on May 25, 2018. The 1.5 oz/Acre rate is off label for a zero day preplant interval and the 3.0 oz/Acre rate is off label altogether. These rates were only used for research purposes to create a worst case scenario and are not encouraged. The 1.0 and 1.5 oz rate were used to rate varietal sensitivity while the 3 oz rate was used to confirm varietal tolerance. Knowledge of sensitivity is important but knowing what varieties are fully tolerant is especially important and that is also why these use rates were chosen. Rainfall was incurred for eight consecutive days after planting totaling 2.68 inches.

Plots were replicated and untreated running checks were present throughout the trial. Ratings were taken at 21 days after application. Ratings were based on degree of stunting and visual leaf injury. Please note that these are the visual results we observed at this location under these conditions. More or less injury could be observed under different conditions. Environmental conditions can have a tremendous impact on the level of observed sensitivity. Consider multiple factors and sources of information when choosing a soybean variety.

Results

Safe	Tolerant
Caution	Moderately Tolerant
Warning	Sensitive
Danger	Highly Sensitive

Table 2: Soybean varietal response to saflufenacil

AGS	**46X17**	Warning
AGS	**48X18**	Warning
AGS	**51X18**	Warning
ASGROW	**45X8**	Caution
ASGROW	**46X6**	Warning
ASGROW	**47X9**	Caution
ASGROW	**48X9**	Caution
ASGROW	**52X9**	Warning
ASGROW	**53X9**	Warning
ASGROW	**56X8**	Safe

ASGROW	**58X9**	Safe
CROPLAN	**RX4555S**	Warning
CROPLAN	**RX4687S**	Caution
CROPLAN	**RX4825**	Warning
CROPLAN	**RX4928**	Danger
CROPLAN	**RX5110**	Warning
CROPLAN	**RX5137**	Caution
CROPLAN	**RX5427**	Danger
CROPLAN	**5548**	Safe
NK	**S45-K5X**	Caution
NK	**S48-R2X**	Caution
NK	**5182X**	Warning
NK	**S56-B7X**	Safe
PIONEER	**42A52X**	Safe
PIONEER	**42A96X**	Warning
PIONEER	**44A72BX**	Safe
PIONEER	**46A57BX**	Safe
PIONEER	**48A60X**	Danger
PIONEER	**49A34X**	Safe
PIONEER	**50A58X**	Safe
PIONEER	**54A75X**	Safe
PIONEER	**55A49X**	Safe
PROGENY	**4620RXS**	Warning
PROGENY	**5016RXS**	Warning
PROGENY	**5688RX**	Caution
USG	**7489XT**	Warning
USG	**7496 XTS**	Danger
USG	**7568XT**	Safe

Conducting Large Scale Drift Study to Demonstrate the Off-Target Movement Potential of New Dicamba Formulation

T. Sandlin

Justification and Methods

Field studies were conducted in North Montgomery County near Deatsville (large field study) and EV Smith REC (low tunnel and particle drift simulation study) in Shorter AL in the summer of 2018.

Deatsville Site: a dicamba tolerant full season soybean variety was planted in May. Application of Xtendimax + Roundup Powermax was made on July 25 10:30am, field soybean height was around 12-14 inch tall. Wind speed was between 3-5 MPH with no sign of temperature inversion. N and NW wind was predominant during application. Nozzle used was TTI 11004 @ 15 GPA. A 10 acre block was sprayed in the middle of this field and sensitive soybean pots were lay out in 10 transects around the sprayed block (figure 1) 30 minutes after application. The soybean pots were kept in sealed trailers up wind to avoid contamination during application. In each transect, 3 soybean pots were put out at 0, 7.5, 15, 25, 50, 100, and 150 ft away from edge of spray block. All pots remained in field site for 48 hours since application and they were watered 3-4 times each day to ensure sufficient moisture. After the initial 48 hours, they were collected back to greenhouse for another 28 days. Visual injury ratings were conducted at 14 and 28 days after application on each pot and averaged for each spot in a transact (table 1 and 2, picture 1 and 2).

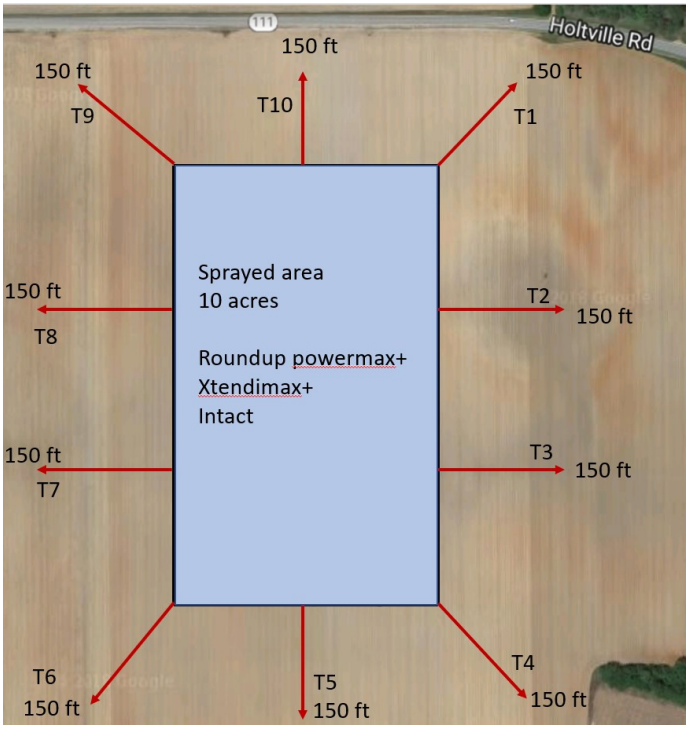


Figure 1. Field map of spray area and sensitive soybean bioassay transacts.

Table 1. Sensitive soybean bioassay averaged injury (%) at 14 days after application.

Transect	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10
0 ft	13	8	10	10	12	7	8	7	10	12
7.5 ft	12	5	10	8	12	8	10	8	5	5
15 ft	7	8	8	5	10	5	8	1	8	7
25 ft	8	5	8	5	7	3	7	2	7	8
50 ft	5	7	3	7	8	3	7	8	5	5
100 ft	0	3	5	5	13	2	-	0	2	3
150 ft	2	3	7	2	3	8	7	0	3	7

Table 2. Sensitive soybean bioassay averaged injury (%) at 28 days after application

Transect	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10
0 ft	17	12	10	10	18	5	17	10	10	10
7.5 ft	13	7	12	8	13	7	10	7	8	10
15 ft	22	7	18	8	22	8	10	5	2	5
25 ft	12	10	15	8	8	7	10	10	10	8
50 ft	8	8	8	8	23	10	13	8	5	10
100 ft	8	13	10	7	13	7	-	5	8	8
150 ft	5	8	5	8	8	12	7	5	7	8

Picture 1: 30% injury on Soybean bioassay (worst injury)

Picture 2: 20% injury on bioassay

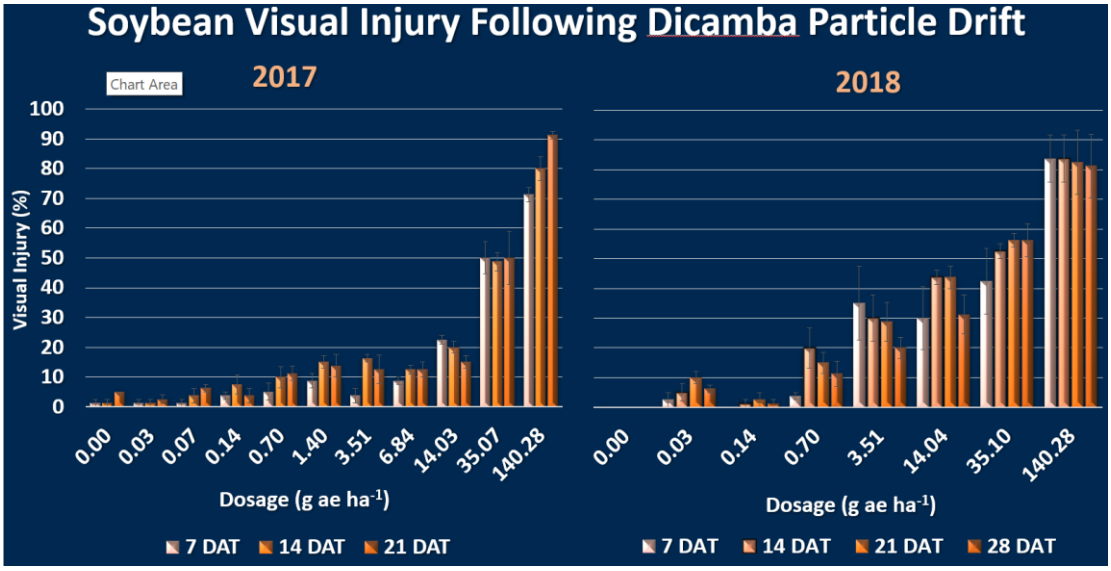
Result summary

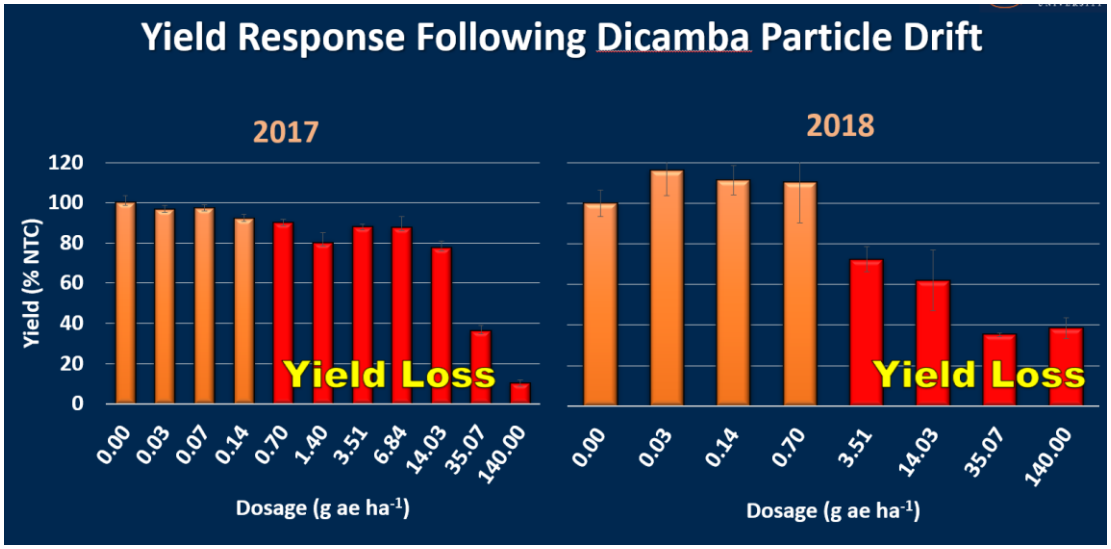
No sensitive soybean bioassay showed more than 30% visual injury caused by dicamba vapor . No particular pattern has been found with soybean damage around the spray block and pots in transects downwind did not show greater damage than pots in other transects. Injury slightly progressed and increased from 14 to 28 days after application, which is consistent from previous

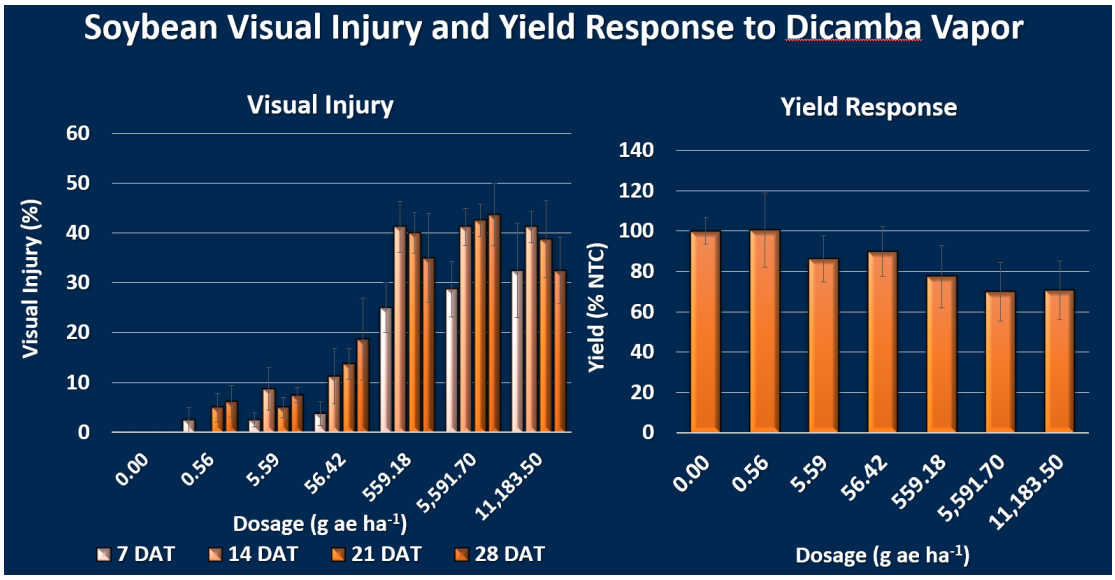
field reports that dicamba vapor damage takes longer time to show on sensitive soybean than particle drift.

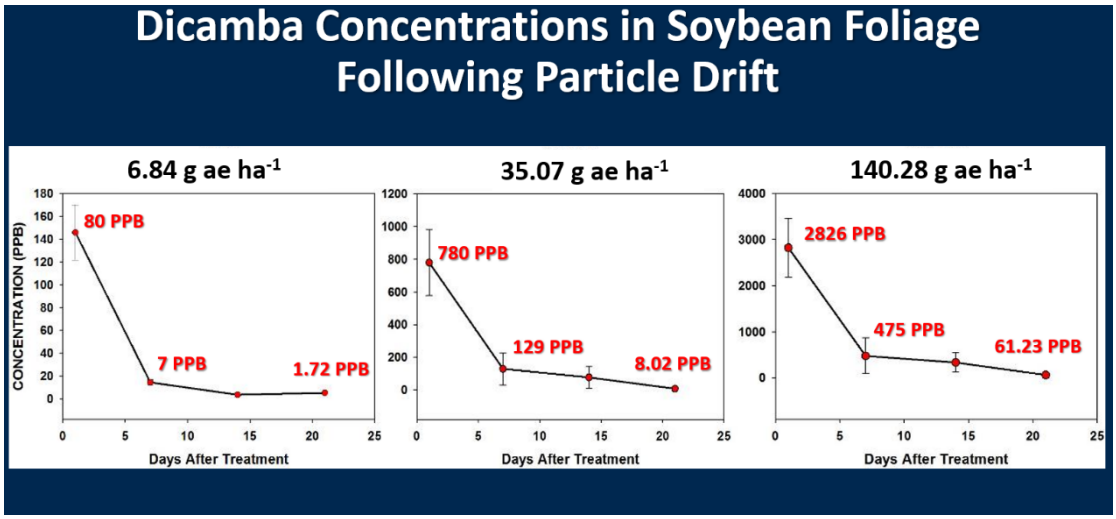
EV Smith site:

Sensitive soybean variety (Pioneer P76T54) was planted at Crop Unit in June of 2017 and 2018. Plot size was 12 x 25 ft (4 rows of soybean in each plot). Experimental design was RCBD with 4 reps. Simulation of particle drift was done with backpack sprayer and handhold boom using TTI110025 nozzles at 15 GPA output. Simulated drift was sprayed on two middles of soybean at R1-R2 stage. Rates used included 0.03 (1/16000th), 0.07 (1/8000th), 0.14 (1/4000th), 0.7 (1/800th), 1.4 (1/400th), 3.51 (1/160th), 6.84, 14.03 (1/40th), 35.07 (1/16th) and 140.3 g ae/ha (1/4th of full label rate) of Xtendimax. For low tunnel study, two soil pans (10 x 7.5 x 2.5 inch) were filled with sandy loam soil and sprayed with 0.56 (1/1000th), 5.56 (1/100th), 56.42 (1/10th), 559 (1X), 5592 (10X) and 11184 g ae/ha (20X of full label rate) of Xtendimax. Then two soil pans were carefully placed in the middle alley of two middle soybean rows, then low tunnel was placed on top of two rows and edges are sealed. Dicamba vapor volatilized from soil pans remained in sealed low tunnels for 48 hours, then low tunnels were removed. Visual injury was evaluated at 1, 3, 7, 14, 21, and 28 DAT. Foliar samples collected from simulated drift study at 1, 14, and 21 DAT. Yield from each plot was also collected at the end of season.




Yield Response Following Dicamba Particle Drift
2017
2018
Yield (% NTC)
120
100
80
60
40
20
0
Yield Loss
Yield Loss
0.00
0.03
0.07
0.14
0.70
1.40
3.51
6.84
14.03
35.07
140.00
Dosage (g ae ha-1)
0.00
0.03
0.14
0.70
3.51
14.03
35.07
140.00
Dosage (g ae ha-1)


Soybean Visual Injury and Yield Response to Dicamba Vapor
Visual Injury
Visual Injury (%)
60
50
40
30
20
10
0
0.00
0.56
5.59
56.42
559.18
5,591.70
11,183.50
Dosage (g ae ha-1)
7 DAT
14 DAT
21 DAT
28 DAT
Yield Response
Yield (% NTC)
140
120
100
80
60
40
20
0
0.00
0.56
5.59
56.42
559.18
5,591.70
11,183.50
Dosage (g ae ha-1)


Dicamba Concentrations in Soybean Foliage Following Particle Drift
6.84 g ae ha-1
CONCENTRATION (PPB)
80 PPB
7 PPB
1.72 PPB
Days After Treatment
35.07 g ae ha-1
780 PPB
129 PPB
8.02 PPB
Days After Treatment
140.28 g ae ha-1
2826 PPB
475 PPB
61.23 PPB
Days After Treatment

Picture 3: Example of low tunnel on two rows of soybean

Result summary

Low dicamba concentrations were detected in the foliage up to 21 DAT. However, data indicated 84-92% of initial concentrations are metabolized within the first 7 days. Soybean visual injury, resulted from particle drift, steadily increased with higher dosages up to 90% for 140.28 g ae ha^{-1}. Compared to the nontreated control, yield losses resulted from particle drift were correlated to dosage with losses of 7% at 0.70 g ae ha^{-1} increasing to 90% at 140.28 g ae ha^{-1}. Alternatively, soybean visual injury resulting from vapor exposure did not exceed 43% regardless of dosage and yield was not significantly reduced. Soybean response to dicamba particle drift was not comparable to vapor exposure. For example, 48-56% injury resulting from particle drift was associated with 64-65% yield reductions in both years. However, 43% injury resulting from vapor exposure was not associated with yield loss. Data suggests visual injury is a poor indicator of yield loss and soybean response is likely to vary with different sources of non-target exposure. Dicamba vapor may injure soybean and unlikely to cause significantly yield loss. Additional research will be needed to fully understand the impact of dicamba drift and vapor damage on sensitive soybeans.

IV. Disease Management

Using Unmanned Aerial Systems (UAS) for Early Detection of Soybean Diseases

E. Sikora, C. Brodbeck, G. Pate, D. Delaney, and A. Hagan

Results for 2018 (year 3 of 3)

In 2016, a preliminary study was launched to examine the potential of using imagery from an unmanned aerial system (UAS) to detect foliar diseases in soybean. Two irrigated fields located at the E. V. Smith Research Center were used for the study. Each trial consisted of replicated plots using two foliar fungicide treatments and an untreated control. Aerial imagery (multi-spectral and true-color) was collected on a biweekly basis during this study. The study was repeated in 2017 with two changes in the outline. First, the study focused only on the 12-acre field allowing for smaller UAS technologies to be tested. Second, a low-cost UAS outfitted with a stock visual camera was utilized to determine if results could be generated comparable to the more expensive fixed wing UAS outfitted with a multi-spectral camera. In 2018 a 12-acre field was also selected and UAS imagery also collected utilizing a multispectral camera.

Using multi-spectral imagery, a Normalized Difference Vegetative Index (NDVI) was generated and compared to direct observations in the field. Also, using the visual camera on the low-cost UAS, a plant health index known as the Visible Atmospherically Corrected Index (VARI) was generated and compared to NDVI imagery, and direct field observations. Disease severity of soybean rust (SBR) was rated on September 18 and October 6th and correlated to UAS imagery collected on Sep 26, Oct. 3 and Oct. 12.

Results from 2017 were similar to those obtained in 2016. SBR was observed in the field on September 18. Significant differences in disease severity between the two fungicide treated programs and the untreated control were recorded on both Sept. 18 and Oct 6th. The more expensive UAS (eBee Plus with Multi-spectral camera) was able to detect the variability caused by SBR as early has Sep. 26, with the variability in the imagery becoming more pronounced in the Oct. 3rd and Oct. 12th images. The low-cost UAS (DJI Phantom 4 with visual camera) was also able to generate imagery which allowed the same variability to be detected. However, it was not until Oct. 3rd that these differences were detected with the low-cost UAS. These differences became more pronounced in the Oct. 12th image with the differences (between treated and untreated plots) for both image sets being significant. SBR had a significant impact on yield with the untreated control yielding 7-10 bu/ac less than the fungicide programs.

In 2018, field during inspections, it was noted that disease was not present. Due to the complete lack of disease, disease ratings were not carried out in the same manner as in 2016 and 2017. After analyzing the UAS imagery, these finding were substantiated with no significant differences being observed in NDVI values between the treatment plots. Furthermore, ni0 significant differences were observed in the yields between the treatment plots.

We have demonstrated during this study that both high and low cost-UAS could be used to detect variability attributed to SBR in soybeans. Disease detection was conducted by assessing variability and identifying either irregular patterns, or patterns that followed the treatments in the study. While UAS is an excellent tool for generating vegetative indices to determine field variability, having a working knowledge of the field, weather patterns, crop stages and management practices is critical for understanding the observed variability. As a conclusion to this study, we feel that a UAS, either outfitted with a multispectral sensor or a simple camera, is a tool that agronomists can keep in their tool chest to utilize as an aide when scouting fields.

Disease and Management of Members of *Fusarium solani* Species Complex Infecting Soybean Fields in Alabama

(Year 1 of a 3-year study)

J. Coleman, A. Pokhrel, K. S. Lawrence, and E. Sikora

Objective

The objectives of this project are to 1) continue to survey field isolates that are responsible for causing soybean sudden death syndrome in Alabama, 2) evaluate previously collected field isolates of the *Fusarium solani* species complex (FSSC) [namely FSSC (3+4) and FSSC 5] and their ability and environmental conditions necessary to cause disease on cultivars of soybean, and 3) assess their management with fungicides.

Results

Last year we reported that the members of the FSSC (3+4) and FSSC 5 were being isolated from diseased soybean roots in Alabama. While this initial analysis relied on multilocus sequencing of the ITS locus and a region encoding the translation elongation factor 1α (*EF-1α*), sequencing and analysis of additional loci encoding other regions used to identify theses fungi [subunits of DNA-directed RNA polymerase II (*RPB1* and *RPB2*)] have revealed that four species within the FSSC were isolated, FSSC (3+4) and FSSC 5, now formerly named as *F. falciforme* and *F. solani*, respectively, as well as *F. paranaense*, and an additional unnamed species of the FSSC.

We have been assessing their virulence on soybean plants in growth chambers and found that of the isolates we have so far assayed, they are virulent on soybean roots and are able to cause root rot. Similar to other root rots caused by members of the FSSC, vascular necrosis initially occurs during the infection on soybean leading to disease development. We plan to continue these assays with the remaining isolates and compare disease development with the FSSC isolates responsible for sudden death syndrome. In addition, we have begun evaluating the susceptibility of these fungi to DMI fungicides, and observed widespread resistance to triadimefon whereas most of the isolates appear susceptible to tebuconazole.

Evaluation of Fungicides for Control of Soybean Rust and Other Foliar Diseases of Soybeans

D. Delaney, E. Sikora and K. S. Lawrence

Objective

To evaluate multiple fungicides for control of soybean rust and other soybean foliar diseases in Alabama.

Results

Fungicide trials for the control of soybean rust (SBR), frogeye leafspot and other foliar soybean diseases were conducted at Tennessee Valley (2), EV Smith-PBU (2) and the Gulf Coast REC (2), for a total of 6 trials. Different trials included different fungicide products, including new ingredients and premixes, timing and varieties. Some trials were irrigated, while most were dryland. One trial was multi-state cooperative, using the same protocol in each state.

Most trials were late planted due to early season wet weather and also intentionally to increase exposure to soybean diseases. However, periods of late summer dry weather at some locations limited disease development until near maturity. Low inoculant levels limited the spread of SBR across the state until late in the season. Frogeye Leaf Spot was common in some trials in 2018; however, weather limited full foliar disease development. Septoria Brown Spot, not usually a major problem in the mid-to-upper canopy in our area, was noticeable this year. Ratings were taken wherever sufficient disease was present.

At Tennessee Valley, Septoria Brown Spot and Frogeye Leaf Spot came in late and ratings were taken. Several treatments were significantly different than the untreated check, and other treatments, for Septoria and Frogeye ratings and soybean yield for the two tests there. Yields ranged from 64 to 75 bu/A for the irrigated test and 41 to 47 bu/A for the dryland trial.

At EV Smith-PBU, Septoria Brown Spot was the primary pathogen (Table 1). Thirteen fungicide treatment combinations and rates were made at the R3 growth stage, in addition to an untreated check, as part of a multi-state protocol. Ratings were taken in early September. All fungicide treatments numerically reduced Septoria ratings, but there were no statistically significant differences between the treatments. There were no significant differences in yield between the check and any of the fungicide treatments, ranging from 42 to 46 bu/A.

At Gulf Coast, Target Spot, Cercospora Leaf Blight, Rhizoctonia and Aerial Web Blight were the major diseases present at sufficient levels to rate. Most fungicides were not very effective in reducing (low) levels of Target spot, Rhizoctonia or Cercospora, although there were some small but significant differences noted. All fungicide applications tested reduced Aerial Web Blight

compared to the Untreated check. Yields ranged from 64-70 bu/A in one trial and, 66 to 74 bu/A in the other, with several fungicide treatments increasing yields 5-8 bu/a compared to the check.

Table 1. Foliar Fungicides for Soybeans at EV Smith-PBU, 2018. Septoria Brown Spot ratings and Yield

Trt. No.	**Treatment**	**Rate**	**Units**	**Septoria Brown Spot (06 Sep)**	**Yield bu/A**
1	Untreated			8.8	44.2
2	Headline	6	FL OZ/A	3.8	46.2
3	Quadris Top SBX	7	FL OZ/A	5.8	44.9
4	Delaro	8	FL OZ/A	3.8	42.6
5	Priaxor	4	FL OZ/A	5.3	45.0
	+ Tilt	4	FL OZ/A		
6	Acropolis	23	FL OZ/A	5.8	41.5
7	Froghorn	20	FL OZ/A	3.8	43.9
8	Domark	4	FL OZ/A	6.3	45.4
9	Topsin 4.5L	20	FL OZ/A	4.0	43.2
10	Topsin 4.5L	20	FL OZ/A	3.3	44.7
	+ Quadris	6	FL OZ/A		
	+ Tilt	4	FL OZ/A		
11	Topsin 4.5L	20	FL OZ/A	4.5	46.0
	+ Quadris	6	FL OZ/A		
	+ Tilt	4	FL OZ/A		
	+ Echo 720	36	FL OZ/A		
12	Revysol (BAS 753)	8	FL OZ/A	3.3	45.3
13	Tilt	6	FL OZ/A	6.3	41.5
			LSD P=.10	*3.65*	*NS*

Evaluation of Fungicide Spray Programs with Large-Scale Strip Tests

E. Sikora, and D. Delaney

We established four large-scale fungicide strip trials at Auburn University research stations to determine the benefit of fungicide applications in soybean production. Trials were established at Belle Mina, Fairhope, Crossville and Shorter. Experiments varied slightly by location but each included an unsprayed control plus single applications of Acropolis, Trivapro (A+B), and Quadris Top SBX. Each trial had a minimum of three replications.

Significant differences among treatments for disease control were only observed at the Fairhope location in 2018. Weather conditions were unfavorable for significant disease development at the other three locations. Results from Fairhope showed a significant benefit of a single fungicide application in controlling frogeye leaf spot, with a significant increase in yield compared the unsprayed control at both locations.

The benefit of a fungicide application is dependent on its timing, with applications made prior to disease onset more effective at protecting the yield potential of the crop. In years when weather conditions do not favor disease development there is rarely a benefit from a fungicide application.

Fairhope

Fungicide/rate	% frogeye leaf spot	Yield bu/A
Untreated control	31.7	45.8
Acropolis 23 oz/a	2.5	53.9
Trivapro A 4 oz/a		
Trivapro B 10.5 oz/a	3.2	53.2
Quadris Top SBX 8 oz/a	3.8	55.1

Fungicide treatments were applied on 08/31/18. Harvest was completed on 11/19/1

In-Vitro Effect of Fungicides on Mycelial Growth of the *Corynespora cassicola* Isolates from Soybean

K. S. Lawrence, J. Koebernick, and M. N. Rondon

Justification

Corynespora cassiicola is a fungal pathogen with importance in soybean producing countries and different genes possibly related to the pathogenicity has been found in isolates sampled from Alabama, known as cassiicolin-encoding genes. Target spot has the *C. cassiicola* as the causal agent, and has been a concern for farms producers and researchers due to its increasing occurrence, causing severity and great damage when not properly controlled. The use of fungicides as chemical control has been a crucial tool to disease management in the agriculture and the diversity of the fungi could be someway acting in the response of these fungicides. For this reason, we expected to explore in vitro the effectiveness of the fungicides over genetically different isolates based on cassiicolin-encoding gene.

Objective

To evaluate the inhibitory effect of different fungicides on mycelial growth of *Corynespora cassiicola* isolates comprehending a different profile of cassiicolin-encoding genes.

Procedures

Sensitivity to fungicides was evaluated based on the mycelial growth of twelve C. cassiicola isolates on potato dextrose agar (PDA) amended with six fungicides concentrations (0.01, 0.1, 0.5, 1, 10 and 100 mg/L of active ingredient) as well as a control, without the addition of fungicide. Fungicide stock suspensions were prepared by dissolving the commercial fungicide, Headline® (pyraclostrobin) and Priaxor® (pyraclostrobin + fluxapyroxad), in sterile deionized water (SDW) prior to use. PDA media amended with the fungicide were poured into plastic Petri dishes. The day after PDA Petri dishes preparation, mycelial plugs (5 mm in diameter) of each isolate from a 10-days-old culture were placed surface down on the center of each Petri dish. All Petri dishes were incubated at 28±2˚C and 12 hours of photoperiod. When one colony in the control treatment (without fungicide) reached the edge of the plate, the mycelial growth (colony diameter) was measured in two perpendicular directions. The diameter of the mycelial plugs was subtracted before calculating the average of the colony and transformed into growth percentage. The experiment was set in a completely randomized design with four replicates per concentration of the fungicide. A Petri dish was used as an experimental unit and the experiment was performed once. EC_{50} values (or fungicides concentration that inhibits 50% of the mycelial growth) for each isolate were estimated by logarithmic regression analysis using SAS 9.4 PROC REG procedure ($P < 0.05$).

Table 1. Fungicides class, active ingredient, product name and rates to evaluate the inhibitory effect on mycelial growth of *Corynespora cassiicola*.

Class	Active ingredient (%)	Product/Trade name	Rate (fl oz/A)
Qol Strobilurins Group 11	Picoxystrobin 22.5%	Aproach 2.08 SC	6.0 - 12.0
Qol Strobilurins Group 11	Fluoxastrobin 40.3%	Evito 480 SC	2.0 - 5.7
Qol Strobilurins Group 11	Pyraclostrobin 23.6%	Headline 2.09 EC/SC	6.0 - 12.0
Qol Strobilurins Group 11	Azoxystrobin 22.9%	Quadris 2.08 SC	6.0 - 15.5
DMI Triazoles Group 3	Cyproconazole 8.9%	Alto 100 SL	2.75 - 5.5
DMI Triazoles Group 3	Prothioconazole 41.0%	Proline 480 SC	2.5-5.0
DMI Triazoles Group 3	Flutriafol 11.8%	Topguard 1.04 SC	7.0 - 14.0
Mixed mode of action	Cyproconazole 7.17% Picoxystrobin 17.94%	Aproach Prima 2.34 SC	5.0 - 6.8
Mixed mode of action	Pyraclostrobin 28.58% Fluxapyroxad 14.33%	Priaxor Xemium	4.0 - 8.0
Mixed mode of action	Azoxystrobin 18.2% Difenoconazole 11.4%	Quadris Top 2.72 SC	8.0 - 14.0
Mixed mode of action	Azoxystrobin 13.5% Propiconazole 11.7%	Quilt Xcel 2.2 SE	10.5 - 21.0
Mixed mode of action	Trifloxystrobin 32.3% Prothioconazole 10.8%	Stratego YLD 4.18 SC	4.0 - 4.65
Mixed mode of action	Bensovindiflupyr 2.9% Azoxystrobin 10.5% Propiconazole 11.9%	Trivapro	13.7-20.7
Mixed mode of action	Fluopyram 15.4% Imadacloprid 22.2%	Velum Total	14.0-18.0
Chloronitriles Group M5	Chlorothalonil 54%	Bravo Weather Stik	16.0-36.0
Dithiocarbamates Group M3	Mancozeb 75%	Manzate Pro Stick	0.75-3.0 lbs/A
MBC Thiophanates Group 1	Thiophanate- methyl 45.0%	Topsin 4.5 FL	10.0 - 20.0
DHI Carboximides Group 7	Penthiopyrad 20.6%	Vertisan 1.67 EC	10.0-30.0
Biofungicide	QST 713 *B. subtillis*	Serenade Opti	14.0-20.0

Amount Requested

Total costs that will be $10,000

1. Salaries
2. Wages
3. Graduate student (1/3) time - $7,500 (Marina Rondon)
    1. Benefits- $ 135
4. Operating
    a. Supplies -$2,000
        i. Petri plates, media, pipets, pots, bags seed, chemicals, gloves and other lab or greenhouse supplies.
        ii. Trips to the PSRC
    b. Travel -$365
        i. Trips to the PSRC

In-Vitro Effect of Fungicides on Mycelial Growth of the *Corynespora cassicola* Isolates

K. S. Lawrence, J. Koebernick, and M. N. Rondon

Results

Twelve *Corynespora cassiicola* isolates sampled from cotton and soybean infected leaves in Alabama with a diversity based on cassiicolin-encoding genes were investigated for sensitivity to two fungicides: QoI fungicide, Headline (pyraclostrobin) and QoI + SDHI fungicide, Priaxor (pyraclostrobin + fluxapyroxad). The concentration of the fungicides ranged between 0.01 – 100 mg/L, and the control treatment without the use of fungicides (Figure 1).

EC_{50} for the fungicide Headline ranged between 17.71 – 66.01 mg/L for *C. cassiicola* isolates from cotton and ranged between 50.03 – 94.50 mg/L for *C. cassiicola* isolates from soybean (Table 1). The mean of the EC_{50} for the fungicide Headline for cotton isolates was 41.89 mg/L, while for soybean isolates was 73.65 mg/L. Higher EC_{50} suggest that these isolates from soybean are less sensitivity to Headline compared to isolates from cotton. Most of the isolates were classified as highly non-sensitive (HNS) to Headline, except for two isolates, BRW03 and EVS01 that were non-sensitive (NS).

EC_{50} for the fungicide Priaxor ranged between 0.57 – 1.03 mg/L for *C. cassiicola* isolates from cotton and ranged between 1.45 – 11.80 mg/L for *C. cassiicola* isolates from soybean. The mean of the EC_{50} for the fungicide Priaxor for cotton isolates was 0.76 mg/L, while for soybean isolates was 5.35 mg/L. Sensitivities of *C. cassiicola* isolates to Priaxor followed the same pattern than sensitivities to Headline, being soybean isolates more sensitive to the fungicides. *C. cassiicola* isolates from cotton were mostly classified as sensitive (S), and just one isolate (FHP01) classified as moderate sensitive (MS). On the other hand, one *C. cassiicola* isolate from soybean was classified as NS, while the rest of the isolates as MS.

Soybean isolates have been more exposed due to the intensive use of these fungicides at the same season or even the long period (years) that these fungicides have been used on soybean fields. Over the years, cotton isolates tend to increase their EC_{50} if the intensive use of the same active ingredients continues. To monitor *C. cassiicola* sensitivity to fungicides is important to manage fungicide resistance and the EC_{50} has been used over the years to represent the loss of sensitivity of different isolates to the fungicides.

Figure 1. *Corynespora cassiicola* mycelial growth in the control treatment (0 mg/L) and the higher concentration (100 mg/L) of the fungicides, Headline and Priaxor.

Table 1. Isolates of *Corynespora cassiicola* and their respective origin and cassiicolin-encoding genes, regression equation, coefficient of determination (R^2), significance (*P-value*), effective concentration of the fungicides Headline and Priaxor (EC_{50}) and sensitivity (S) to the fungicide.

Isolate	Origin	Gene	Headline (pyraclostrobin)					Priaxor (pyraclostrobin + fluxapyroxad)				
			Equation [z]	R^2	*P-value*	EC_{50} (mg/L)	S [y]	Equation	R^2	*P-value*	EC_{50} (mg/L)	S [x]
BRW03	Cotton	*Cas2*	$y = 66.712\ e^{-0.015x}$	0.79	<.0001	19.29	NS	$y = 88.191\ e^{-0.929x}$	0.94	<.0001	0.61	S
EVS01	Cotton	*Cas2*	$y = 79.090\ e^{-0.026x}$	0.21	0.0015	17.71	NS	$y = 90.028\ e^{-0.882x}$	0.94	<.0001	0.67	S
FHP01	Cotton	Cas0	$y = 74.306\ e^{-0.008x}$	0.69	<.0001	49.96	HNS	$y = 66.092\ e^{-0.270x}$	0.75	<.0001	1.03	MS
FHP22	Cotton	*Cas2*	$y = 85.683\ e^{-0.008x}$	0.86	<.0001	66.01	HNS	$y = 85.541\ e^{-0.949x}$	0.92	<.0001	0.57	S
HSV01	Cotton	*Cas2*	$y = 77.683\ e^{-0.008x}$	0.75	<.0001	55.84	HNS	$y = 66.841\ e^{-0.419x}$	0.91	<.0001	0.73	S
HSV12	Cotton	Cas0	$y = 74.939\ e^{-0.010x}$	0.82	0.0002	42.55	HNS	$y = 66.815\ e^{-0.312x}$	0.81	<.0001	0.93	S
LIM02	Soybean	*Cas2*	$y = 77.461\ e^{-0.009x}$	0.79	0.0059	50.03	HNS	$y = 71.705\ e^{-0.161x}$	0.78	<.0001	2.24	MS
LIM13	Soybean	Cas0	$y = 79.702\ e^{-0.009x}$	0.87	<.0001	51.81	HNS	$y = 74.638\ e^{-0.181x}$	0.83	<.0001	2.22	MS
LIM14	Soybean	*Cas6*	$y = 94.979\ e^{-0.007x}$	0.92	<.0001	94.50	HNS	$y = 86.393\ e^{-0.378x}$	0.99	<.0001	1.45	MS
PBU04	Soybean	*Cas2+6*	$y = 94.812\ e^{-0.008x}$	0.94	0.0003	83.21	HNS	$y = 57.100\ e^{-0.021x}$	0.54	<.0001	6.27	MS
PBU06	Soybean	*Cas2*	$y = 94.919\ e^{-0.007x}$	0.90	<.0001	90.28	HNS	$y = 64.857\ e^{-0.032x}$	0.66	<.0001	8.13	MS
PBU07	Soybean	*Cas6*	$y = 89.570\ e^{-0.008x}$	0.92	<.0001	72.06	HNS	$y = 65.013\ e^{-0.022x}$	0.75	<.0001	11.80	NS

[z] y represents the percentage of mycelial growth inhibition; x represents the fungicide concentration.

[y] Sensitivity (S) of *Corynespora cassiicola* to QoI fungicide according to Teramoto et al. (2017): S = sensitive (< 0.16 mg/L); MS = moderate sensitive (0.16 – 1.0 mg/L); NS = non-sensitive (1 – 25 mg/L); HNS = highly non-sensitive (> 25 mg/L).

[x] Sensitivity (S) of *Corynespora cassiicola* to SDHI fungicide according to Teramoto et al. (2017): S = sensitive (< 1.0 mg/L); MS = moderate sensitive (1 – 10 mg/L); NS = non-sensitive (10 – 25 mg/L); HNS = highly non-sensitive (> 25 mg/L).

Development of Reliable Screening Method for Resistant Varieties of Soybeans to Target Spot

K. S. Lawrence, J. Koebernick, and M. N. Rondon

Objective

Determine a high throughput method to screening soybeans varieties to target spot.

Leaf-wilting bioassay – This method will be set up with the immersion of a trifoliate soybean leaves in the culture filtrate containing crude toxin. The culture filtrate will be obtained as beforehand described. The petioles of trifoliate leaves will be immediately immersed in 5 ml of the culture filtrate in small tubes. As control, trifoliate leaves will be immersed in 5 mL of distilled water and non-inoculated PDB medium. According to Fernando et al (2010), the degree of wilting will be grouped into three categories as mild, moderate or fresh water / dry weight in percent of a control (Breton et al., 2000).

Toothpick-inoculation method – This method will be set up with the inoculation of a single toothpick previously immersed in the crude toxin. Soybean varieties and breeding lines will be growth at the PSRC greenhouse. Ten soybean seeds will be sown per pot and 14 days after planting, a single toothpick will be inserted into the stem between the cotyledons and the first true soybean leaf. Plants will be maintained under high humidity (> 80%) for 48 hours. The incidence of plants with wilting symptoms will be rated daily for 10 days. From the multiple disease incidence ratings, the area under disease progress curve (AUDPC) will be calculated.

Leaf-puncture bioassay – This method will be set up by placing drops of the crude toxin on the adaxial side of detached soybean leaves. Detached leaves of soybean will be placed in Petri dishes in moist conditions and inoculated with 20 µL drops of toxin. As control, detached leaves will be treated with distilled water and non-inoculated PDB medium. Petri dishes will be incubated at 25˚C, photoperiod 12h until the development of the symptoms. Lesions will be grouped using the rating system: group I – no reaction; group II, pin point sized lesions; group III, pin head sized lesions; group IV, moderately extended lesions with slight growth of mycelium; group V, more extended lesions with profusely grown mycelium. The symptoms intensity (SI) will be expressed as the mean lesion area ± the standard error from the 10 inoculated leaves (six leaves per inoculation and two biological replicates (Breton et al., 2000; Fernando et al., 2010).

Detached-leaf bioassay – This method will be set up by placing drops of the conidia suspension on the adaxial side of detached soybean leaves. The *C. cassiicola* conidia will be adjust to $2x10^4$ conidia/mL. Detached leaves of soybean will be placed in Petri dishes in moist conditions and inoculated with 20 µL drops of the conidia suspension. As control, detached leaves will be treated

with distilled water and non-inoculated PDB medium. Petri dishes will be incubated at 25˚C, photoperiod 12h until the development of the symptoms. Lesions and the symptoms intensity will be evaluated as previously described.

Plant inoculation – This method will be conducted at the PSRC greenhouse and five soybean seeds will be sown per pot, and thinned to two planta per pot seven days after planting (DAP). The *C. cassiicola* isolates will be cultivated and conidia will be collected and adjust to $2x10^4$ conidia/mL. The conidia suspension will be supplemented with 0.02% Tween20 for inoculation which will be applied to both the adaxial and abaxial leaves of 30 days-old soybean plants until runoff. After inoculation, plants will be covered with transparent plastic bags for 72 hours and will be kept in the greenhouse for the duration of the trial. Symptoms will be scored 10 days after inoculation using the disease rating system developed by Onesirosan et al. (1973): 0, no lesions on leaves or stems (no symptom); 1, weakly virulent or hypersensitive response: a few to many nonexpanding pinpoint lesions; 2, moderately virulent: many expanding lesions, some coalescing, but not resulting in blight; and 3, highly virulent: lesions spreading to form large areas of dead tissue resulting in a blighting effect. The trial will be arranged in a randomized complete block design (RCBD) with five replications and will be repeated at least once.

Outcome

Our findings will help assess soybean varieties and breeding lines to *C. cassiicola* based on multiple plant assays to enhance the soybean breeding program.

Amount Requested

Total costs that will be $10,000

1. Salaries
2. Wages
3. Graduate student (1/3) time - $7,500 (Marina Rondon)
    1. Benefits- $ 135
4. Operating
    a. Supplies -$2,000
        i. Petri plates, media, pipets, pots, bags seed, chemicals, gloves and other lab or greenhouse supplies.
        ii. Trips to the PSRC
    b. Travel -$365
        i. Trips to the PSRC

Development of Reliable Screening Method for Resistant Varieties of Soybeans to Target Spot

K. S. Lawrence, J. Koebernick, and M. N. Rondon

Target spot tolerant germplasm seeds of soybean were requested in 29/March/2018 from the U.S. National Plant Germplasm Resources, GRIN-Global (https://npgsweb.ars-grin.gov/) to conduct these trials. Only 50 seeds of each germplasm requested were obtained which affected our plan for the trials, and an unexpected seed multiplication were conducted from May-October/2018 in microplots located at PSRC in Auburn, AL.

After seeds multiplication, trials with leaf-wilting bioassay methodology were started using two soybean germplasms: susceptible (NKS 56-B7X) and tolerant (RA-606). Four replicates of trifoliate soybean leaves of each germplasm were immersed in a solution containing 50% of distilled water and 50% of culture filtrate of different isolates and the result recorded as level of wilting (1 – mild, 2 – moderate, 3 – severe).

Results

Trial 1 – Conducted with six culture filtrates of isolates previously sampled from soybean plants with a diversity based on cassiicolin-encoding genes, and water as the negative control. All variables were statistically significant ($P < 0.05$). The susceptible soybean germplasm exhibited higher wilting of soybean leaves compared to tolerant soybean germplasm ($P < 0.05$). The choice of the germplasm it is an important factor when developing a leaf-wilting bioassay, taking into the consideration that it is necessary to have a positive control (susceptible germplasm) when screening a wide number of genotypes.

PBU06 (*Cas2*) and LIM13 (Cas0) were the isolates that resulted in the higher wilting of soybean leaves ($P < 0.05$). The absence of cassiicolin-encoding gene (Cas0) did not affect the toxicity of the culture filtrate obtained from the isolate LIM13 to the soybean leaves tested. Other substances, metabolites or toxins may be produced by isolates with the absence of cassiicolin-encoding genes that are responsible for inducing soybean leaves wilting. Different culture filtrates (LIM02, LIM14, PBU04, PBU07) exhibited a similar behavior compared to the negative control (water) ($P < 0.05$), even though these culture filtrates were obtained from isolates with different cassiicolin-encoding genes. These findings suggests that some isolates are not toxic enough to be used as an indicator of soybean leaves wilting.

Trial 2 – Conducted with six culture filtrates of isolates previously sampled from cotton plants with a diversity based on cassiicolin-encoding genes, two of the most toxic culture filtrates of isolates from soybean tested in the first trial (LIM13 and PBU06), and water as the negative control. Only culture filtrates were statistically significant ($P < 0.05$). PBU06 (*Cas2*) was the isolate with higher wilting of

soybean leaves, followed by FHP01 (Cas0) ($P < 0.05$). Most of the isolates exhibited lower induction of soybean leaves wilting, similarly to the negative control (water) ($P < 0.05$). Results demonstrated that independently of the soybean germplasm reaction to target spot (susceptible or tolerant), the isolates choice for screening soybean germplasm using leaf-wilting bioassay could affect the results. The use of culture filtrates from cotton isolates may not have an effect over soybean leaves wilting, indicating some host-specificity. The presence or absence of cassiicolin-encoding genes did not exhibited a high influence on the soybean leaves wilting.

Screening of soybean germplasm can be facilitated using leaf-wilting bioassay because is a non-destructive method. Regarding the methodology used to screen soybean germplasm, it is important to know the pathogen variability and their specificity to the host.

Table 2. Soybean leaf wilting for two trials using culture filtrates of different *Corynespora cassiicola* isolates.

TRIAL 1					**TRIAL 2**				
Source of variation			***df***	***P-value*[z]**	Source of variation			***df***	***P-value***
Cultivar			1	<.0001	Cultivar			1	0.0816
Culture Filtrate			6	<.0001	Culture Filtrate			8	0.0021
Cultivar x Culture Filtrate			6	<.0001	Cultivar x Culture Filtrate			8	0.9800
Cultivar LS-means					Cultivar LS-means				
Susceptible (NKS 56-B7X)			1.57	a	Susceptible (NKS 56-B7X)			1.64	a
Tolerant (RA-606)			1.00	b	Tolerant (RA-606)			1.36	a
Culture Filtrate LS-means					Culture Filtrate LS-means				
Isolate	Origin	Gene			Isolate	Origin	Gene		
LIM02	Soybean	*Cas2*	1.00	b	BRW03	Cotton	*Cas2*	1.00	c
LIM13	Soybean	Cas0	1.88	a	EVS01	Cotton	*Cas2*	1.00	c
LIM14	Soybean	*Cas6*	1.00	b	FHP01	Cotton	Cas0	2.00	ab
PBU04	Soybean	*Cas2+6*	1.13	b	FHP22	Cotton	*Cas2*	1.50	bc
PBU06	Soybean	*Cas2*	2.00	a	HSV01	Cotton	*Cas2*	1.25	c
PBU07	Soybean	*Cas6*	1.00	b	HSV12	Cotton	Cas0	1.38	bc
Control (water)			1.00	b	LIM13	Soybean	Cas0	1.50	bc
					PBU06	Soybean	*Cas2*	2.38	a
					Control (water)			1.50	bc

[z] LS-means of wilting followed by the same letter(s) within a column are not statistically different based on Fisher's Protected LSD ($P < 0.05$).

Evaluation of Biological Control Agents' Potential to Cause Systemic Resistance in Soybean

K. S. Lawrence, E. Sikora, and K. Gattoni

Justification

Meloidogyne incognita, the southern root-knot nematode is an important pathogen of soybeans. One management strategy for *M. incognita* is the use of biological control agents. Various species of *Bacillus* sp., distinguished as plant growth-promoting rhizobacteria, have exhibited potential as biological control agents against *M. incognita*. Biological control agents work by either releasing metabolites after colonizing the plant roots or by systemic resistance, which will activate and increase the response by the plant's defense system. Systemic resistance can occur by the upregulation of two different pathways, one utilizing salicylic acid and the other utilizing jasmonic acid. Determining the mechanism by which biological control agents act in regards to nematode control is essential in the further utilization and potential commercialization of each agent. Therefore, the main goal of this project is to determine whether *Bacillus* species can prompt a systemic response to *M. incognita* in soybeans.

Objective

The main objective of this project is to determine if five *Bacillus* sp. systemically induce plant defenses in soybean to defend against *M. incognita*. An in vitro assay will be performed to look at the direct interaction between the *Bacillus* sp. and *M. incognita* second stage juveniles. A split root assay will determine the potential for each *Bacillus* sp. to induce systemic resistance by placing *M. incognita* and the *Bacillus* sp. on separate root halves of the same soybean plant. Finally, an RT-qPCR will be done to look at genes correlated to the upregulation of jasmonic and salicylic acid to confirm systemic resistance.

Results

The first experiment performed was a greenhouse pot test. Soybean seeds were planted and inoculated with 5,000 *M. incognita* eggs per mL and one of the following treatments; 1) control, 2) Fluopyram 3) *B. firmus* I-1582, 4) *B. amyloliquedfaciens* QST713, 5) *B. pumilus* GB34, 6) *B. velenzensis* strain 2, and 7) *B. mojavensis* strain 3. All bacteria were inoculated at a volume of 1mL per seed and at a concentration of 1 x 10^6 cfu/mL. After 30 days the plant parameter were measured and *M. incognita* eggs were extracted. The results of this showed a statistical difference in plant height between the treatments. *Bacillus firmus* I-1582, *B. pumilis* GB24, and *B. mojavensis* strain 3 were able to significantly increase the plant height compared to the chemical control of Fluopyram (Table 1). Similarly, Fluopyram had the lowest, numeric, biomass, which is the root

fresh weight plus the shoot fresh weight, and *B. firmus* I-1582 and *B. mojavensis* strain 3 had the highest, numeric, biomass (Table 1). Fluopyram had the lowest, numerically, number of *M. incognita* eggs per gram of root, followed by *B. amyloliquefaciens* QST713 (Table 1). The other *Bacillus* treatments decreased, numerically, the number of *M. incognita* eggs per gram of root compared to the control, but this was not significant (Table 1).

Tables 1: Plant height (mm), biomass (g) and *M. incognita* eggs per gram of root obtained from the greenhouse pot test ($p \leq 0.1$). This data represents one repetition of the test.

Treatment	Plant Height	Biomass	RKN eggs per gram of root
Control	32.6 ab	7.55	302.03
Fluopyram	26.72 b	5.44	75.02
B. firmus I-1582	35.84 a	8.24	252.92
B. amyloliquefaciens QST713	32.39 ab	7.52	136.47
B. pumilis GB34	34.06 a	7.90	219.15
B. velenzensis strain 2	33.21 ab	7.88	205.74
B. mojavensis strain 3	33.96 a	8.26	213.00

The in vitro assay determined the percent mortality of *M. incognita* second stage juveniles caused by the select *Bacillus* sp. and their metabolites. The *Bacillus* sp., a water control and the *Bacillus* sp. metabolites, extracted according to Apetroaie-Constantin et al., 2008, were added to each well of a 96 well plate along with a approximately 30 *M. incognita* second satge juveniles. The *Bacillus* sp. were added at a concentration of 1×10^6 cfu/mL. The percent mortality was determined using the number of living and dead nematode at 0 hours and 48 hours. Fluopyram was not used in this assay because it is a thick, opaque white liquid that made percent mortality determination difficult. After 48 hours the, *B. firmus* I-1582, the *B. firmus* I-1582 metabolites and the *B. amyloliquefaciens* QST713 increased the percent mortality compared to the water control (Fig 1). This indicated that *B. firmus* I-1582 can directly antagonize the nematode, potentially through the release of a metabolites. It is unknown whether *B. amylolieufaciens* QST713 can release the metabolites that caused an increase in percent mortality (Fig 1), therefore it cannot be concluded that this *Bacillus* sp. works by direct antagonism. This test was also done on the soybean cyst nematode, *Heterodera glycines*, but none of the treatments were able to increase percent mortality compared to the control.

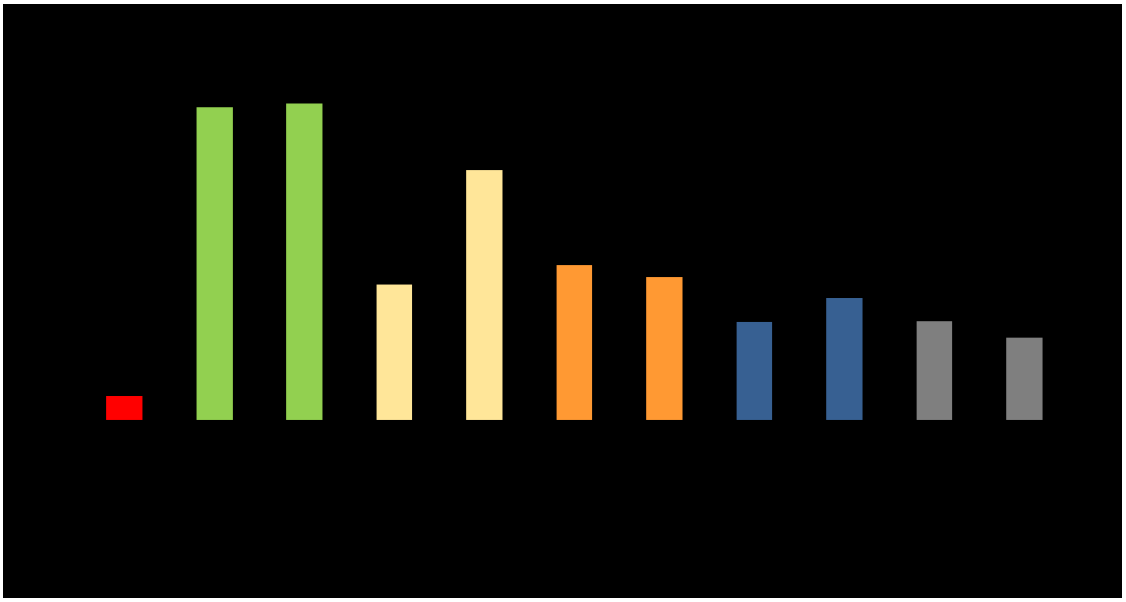


Figure 1: Percent mortality obtained from the in vitro assay using the intact bacteria and *Bacillus* sp. metabolites ($p \leq 0.05$). This data represents three repetitions of the assay.

The final assay performed was a split root assay. This assay was performed by initially germinating soybean seeds in germination paper for between 4-6 days and then cutting the end 1 mm of the small root and planting that in a sand and fertilizer mixture. The plant was left to grow for between 1 and 2 weeks or until 2 equal root halves were developed. The root haves were placed in separate conetainers positioned immediately next to each other with a small plastic cup with the bottom cut off positioned evenly between the conetainers, holding the shoot. There were five patterns of inoculation for each treatment including: 1) a control with no inoculation on either root half (control), 2) bacteria or fluopyram inoculated on root half A and no inoculation on root half B (bacteria control), 3) no inoculation on root half A and *M. incognita* eggs inoculated on root half B (nematode control), 4) bacteria or fluopyram and *M. incognita* eggs inoculated on root half A and no inoculation on root half B, and 5) bacteria or fluopyram inoculated on root half A and *M. incognita* eggs inoculated on root half B. The soybeans were inoculated with the same volume and concentration of bacteria and nematode eggs as described in the greenhouse pot test. The results of this assay showed no difference in plant height or biomass between any of the treatments. Fluopyram when inoculated concomitantly with the nematode was the only treatment able to decrease the nematode eggs per gram of root significantly (Table 2). *Bacillus amyloliquefaciens* QST713 was able to decrease the eggs per gram of root, numerically, when inoculated concomitantly and when inoculated on the same side as the nematode, indicating that this was the most successful *Bacillus* sp. treatment (Table 2). *Bacillus velenzensis* strain 2 followed by *B.*

firmus I-1582 seemed to be the next best *Bacillus* sp. treatments (Table 2). Since these three treatments were the most successful, they will be analyzed in the RT-qPCR.

Table 2: Plant height (mm), biomass (g) and *M. incognita* eggs per gram of root obtained from the split root assay ($p \leq 0.1$). This data represents three repetitions of the assay.

Treatment	Plant Height	Biomass	RKN eggs per gram of root
Control(A) Control(B)	28	12	NA
Control(A) *M. incognita*(B)	26	12	888 a
B. velenzensis strain 2(A) Control (B)	25	10	NA
B. velenzensis+*M. incognita* (A) Control (B)	29	12	673 a
B. velenzensis (A) *M. incognita* (B)	28	11	554 a
B. firmus I-1582(A) Control (B)	29	11	NA
B. firmus+*M. incognita* (A) Control (B)	29	11	943 a
B. firmus (A) *M. incognita*(B)	26	11	514 ab
B. pumilis GB34(A) Control (B)	28	11	NA
B. pumilis+*M. incognita* (A) Control (B)	25	12	958 a
B. pumilis (A) *M. incognita* (B)	29	14	603 a
B. amyloliquefaciens QST713(A) Control (B)	27	11	NA
B. amyloliquefaciens+*M. incognita* (A) Control (B)	26	12	596 a
B. amyloliquefaciens (A) *M. incognita* (B)	28	10	551 ab
B. mojavensis strain 3(A) Control (B)	29	11	NA
B. mojavensis+*M. incognita* (A) Control (B)	29	14	813 a
B. mojavensis (A) *M. incognita* (B)	28	12	627 a
Fluopyram (A) Control (B)	25	11	NA
Fluopyram+*M. incognita* (A) Control (B)	24	9	277 b
Fluopyram (A) *M. incognita*(B)	28	11	466 ab

An RT-qPCR assay has begun to be analyzed. Soybean plants at the second true leaf stage were inoculated with the chosen *Bacillus* sp. and *M. incognita* second stage juveniles and samples were collected at 0 hours (h), 1h, 3h, 6h, 12h, 24h, 48h, 96 h, and 1 week after inoculation. The RNA was extracted with the Sigma Spectrum™ Plant Total RNA Kit. The cDNA will be synthesized with the Goscrpit™ Reverse Transcription System Kit and the Rt-q-PCR will be performed using PerfeCTA® SYBR® Green Fastmix® ROX qPCR Master Mix.

Outcome

Our findings will help assess ability of *Bacillus* species to produce systemic induced resistance to *M. incognita* and how best to utilize these biological control agents in a commercial setting.

Building a Disease-Related Gene Expression Catalog that Can be Used for Disease Diagnosis and Genetic Improvement

B. Locy, E. Sikora, K. Conner, A. Rashotte, and K. S. Lawrence

Any delay in diagnosing a plant disease disorder can result in a grower making an unnecessary pesticide application due to the uncertainty of the causal agent. Unfortunately, soybean diseases early in their development can be difficult to differentiate in the field or laboratory. This was evident in 2015 when charcoal rot, stem canker and sudden death syndrome, three diseases with similar symptoms early in their progression, were active in soybean fields in Alabama. Having a rapid disease diagnostic kit available to the diagnostician that could identify a disease before symptoms are expressed would provide critical time for a grower to respond to a disorder. This project involves preliminary studies aimed at providing proof of concept that nanotechnology-based disease diagnostic sensors could be developed for a myriad of soil-born diseases in soybean.

Objective

Our objective is to establish a soybean disease-related gene expression catalog (SDGEC). This has been done using next generation DNA sequencing techniques to identify all specific messenger RNAs (mRNAs) found in plant tissues, not infected with (control) or infected with pathogens (infected). A gene expression catalog should contain sequences derived from tisssues infected with a range of pathogens so that gene expression patterns can be compared for different pathogens. At the present time we have built an SDGEC catalog for control and for plants infected with 3 nematode species, i.e. reniform nematode (ren), root knot nematode (rkn), and soybean cyst nematode (scn). Beside the sequencing, the raw sequence data from the Hudson-Alpha Institue in Huntsville, AL must be assembled into a transcriptome database, and the database must be documented as to quality of the assembled sequences. At the present time our transcript assembly (database) contains 218,919 assembled transcripts (mRNAs derived from genes). (Note that this assembly also contains sequences from the southern blight fungal samples discussed below.) Our assembly covers 156,443,037 nucleotides and has an average transcript length of 715 nucleotides. Fifty percent of the sequences are at least 1105 nucleotides long, and more than 95% of the sequences can be found in the soybean genome. Circa 70% of the sequences cover at least 70% of the corresponding soybean gene (from the genome). These general statistics indicate that our assembly is a reasonably high quality.

We have also built or are building a catalog entries for 3 fungal pathogens, i.e. southern blight (SB), charcoal rot (CR), and sudden death syndrome (SD). The SB entry is complete, and the assembly statistics are included in the data given above. The CR and SD samples (with appropriate

controls) have been prepared; RNA has been extracted; and these samples are awaiting sequencing. Before we can proceed, we expect to complete sequencing with the remaining two diseases within 2 months, and then an additional 2 months will be required to assemble the raw sequence, verify the assembled sequence, and have it ready for analysis.

While we are completing building our SDBEC, we have proceeded to analyze the entries we already have for the three nematode species above.

To date, progress on this effort has successfully identified changes in mRNA expression in leaves and roots during reniform nematode (ren) infestation, root knot nematode (rkn) infestation, and soybean cyst nematode (scn) infection. The objective of this phase of the work is to identify sequences that are up-regulated in soybean leaves, when each of the nematodes infect roots. These results of the preliminary analysis are summarized in the table below:

Table 1. Isoform analysis of the soybean transcripts differentially expressed in leaves and roots of plants infected with reniform, root knot, and soybean cyst nematode.

	Total Genes Expressed	Total Differential Expressed Genes (FDR<0.005)	DOWN-regulated	UP-regulated	Down-regualted (absolute)	UP-regualted (absolute)	Unique
Reniform-Leaf	55,511	166	66	77	53	48	**45**
Reniform-Root	54,417	171	59	112	41	52	
Root Knot-Leaf	38,784	620	111	509	26	46	**35**
Root Knot-root	47,410	488	105	383	52	141	
SoyCyst-Leaf	37,562	357	201	156	59	23	**14**
SoyCyst-Root	47,109	358	124	234	68	27	

The second column in the table indicates the total number of expressed genes in each of the samples, while the 3rd column shows the total number of these genes that are expressed differently in tissues of pathogen infected plants compared to uninfected control plants (DE). Particularly note that there are hundreds of DE isoforms (genes) expressed in leaves of the plants that have a root-born nematode infection (gray squares). The next two columns report the number of genes that are down and up regulated, respectively. Down- regulated means genes whose expression goes down upon pathogen infection, while up-regulated means genes whose expression goes up upon pathogen infection. Again note in the green boxes that there are many up-regulated genes available to us that could serve as distinctive markers for nematode infection. However, the most useful potential markers would be those that are expressed only in the infected plant tissues and not in uninfected plant tissues. These are referred to as absolutely down- or up-regulated

transcripts. Note in the yellow boxes that there are between 23 and 48 transcripts that fall into this absolute category, meaning they are only expressed in nematode infected tissues. The right-most column shows how many of the potential up-regulated signature sequences (yellow boxes, and column) are specific/unique to the pathogen involved (blue boxes on right). These sequences potentially may be useful as leaf pathogen specific signature sequences that can be used to critically identify pathogen infection of the roots by sampling leaves (the objective of this phase of the work).

However, to verify the utility of these potential sequences it is necessary to analyze them further. The first thing we did was to examine the nature of the sequences. It turns out that there are a number of nearly identical sequences (called isoforms) that are found in the unique sequences (blue boxes in Table 1). This means that while there is a transcript that is only expressed in a particular tissue, other variants that may differ by as little as one nucleotide (out of perhaps thousands) are found in the isoform analysis given in Table 1. Such isoforms complicate our analysis and make identifying useful signature sequences difficult. Accordingly, we have done a parallel gene (rather than isoform) analysis using the same sequence data as for table 1. Genes in this situation means that all transcript isoforms of a specific transcript are grouped into a gene. As such the number of genes is less than the number of isoforms, but all highly similar sequences score as one gene rather than multiple isoforms. The results of the gene analysis are given in Table 2 below:

Table 1. Isoform analysis of the soybean transcripts differentially expressed in leaves and roots of plants infected with reniform, root knot, and soybean cyst nematode.

	Total Genes Expressed	Total Differential Expressed Genes (FDR<0.005)	DOWN-regulated	UP-regulated	Down-regualted (absolute)	UP-regualted (absolute)	Unique
Reniform-Leaf	52,646	327	233	94	1	3	3
Reniform-Root	51,597	205	32	173	3	2	
Root Knot-Leaf	37,220	1,203	271	921	0	6	4
Root Knot-root	45,444	3,718	1277	2441	0	61	
SoyCyst-Leaf	36,180	729	385	344	15	5	2
SoyCyst-Root	45,232	1,075	211	854	1	1	

This table shows that there are 3, 4, and 2 sequences that can be used for monitoring reniform, root knot, and soybean cyst nematode, respectively. These appear to be specific/unique sequences expressed in leaves of infected plants in response to each of these 3 nematodes. We are preparing

to assay these sequences using PCR and moving forward with that tool for assessment of plants from the field in the coming season.

The analysis of our current SDEGC for the gene expression patterns following infection of the 3 types of nematodes will also yield valuable information about how each of these pathogenic nematodes work. Such information may well reveal novel approaches for nematode control and can be a valuable as a tool in a breeding program for nematode resistance. The details of this full transcriptome analysis are being presented in the Biochemistry & Biotechnology section at the Southern Association of Agricultural Scientists taking place in Birmingham, AL on Feb 4, 2019. And a paper describing the details of this analysis is in preparation.

System Biology of Plant-Growth Promoting Rhizobacteria (PGPR)-Induced Drought Tolerance in Soybean

S. W. Park and E. Sikora

Objective

A long-term goal of our laboratories is, employing generic engineering and molecular breeding approaches, to develop new drought tolerance lines of soybean. However, the major and present obstacle is that most - if not all - known drought tolerance (responsive) genes are not usable for our goal, because those genes mostly engender the stomata closure, and consequently the growth suppression of plants once ectopi- cally expressed. Hence, the proposed study has aimed at exploring novel candidate genes and genetic reper- toire that function in the activation of drought tolerance but are not involved in stomata closure processes in plants.

Rationale and Significance

Our earlier studies demonstrated that cohabitation of soil-borne PGPR such as *Paenibacillus polymyxa* can induce (**prime**) drought tolerance and concurrently growth promotion in soy- beans (Fig. 1). These results suggest that the inoculation of *P. polymyxa* enables soybeans to activate the expression of certain genes, which **a**) heightens their states of tolerance against water deficiency, and **b**) pro-motes their growth and development.

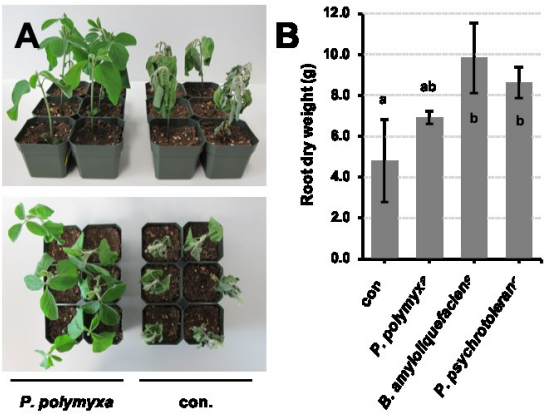


Figure 1. *P. polymyxa* is able to prime drought toleran and promote growth in soybeans. **A.** Root inoculation *P. polymyxa* (2x 10ml of 10^8 cfu•ml^{-1}) engenders droug tolerance of Soybeans. The photographs were taken at wk after stopping watering. **B.** Root inoculations of PGF (2x10ml of 10^8 cfu•ml^{-1}) including *P. polymyxa* promo root growth, measured by the dry weights (g) of soybe roots collected at 2-wk post PGPR inoculations. Note th *Bacillus amyloliquefacience* and *P. psychrotolerances* a negative controls of our study, incapable of priming drought tolerance in soybeans

Therefore, identification and characterization of gene induced by *P. polymyxa*-induced genes that are capable of priming drought tolerance without, if not enhance, compromising growth will provide new gen- etic and biotechnological resources to solve one of the most eminent and immediate environmental chal- lenges in the world, drought and global warming.

Progresses

We have used the high-resolution, quanti- tative (q)RT PCR analyses, and surveyed *P. polymyxa*-responsive gene expressions. Results from these studies have revealed that;

1. *P. polymyxa* control selective abscisic acid (ABA) signaling-responsive genes (ARGs, e.g.,

RD29A) (Fig. 2). Note that ABA signaling plays crucial roles in the activation of drought tolerance.

2. *RD29A* (ABA-responsive gene) & *RD29B* (drought- responsive gene) are *P. polymyxa*-inducible **memory** genes. The memory genes are known to be produced in considerably higher transcript levels during one or more subsequent stress, relative to the initial encounter with the stress; genes responding similarly to each stress form the 'non-memory' category (BMC Plant Biol 13: 229).

For instance, the first inoculation of *P. polymyxa* induced increased expressions (~5-folds) of *RD29A* and *RD29B*, while the second-time inoculation of *P. polymyxa* at 48-hr post the primary *P. polymyxa* inoculation(hp1°i) led to even greater induction of both genes at 78 hp1°i, in comparison to those after the primary inoc- ulation (e.g., 6 and 30 hp1°i; Fig. 2). Importantly, the expression of *RD29A* and *RD29B* is **a**) regulated rhythmically by the diurnal cycle (called circadian control, Fig. 2), and **b**) unresponsive to negative control PGPR such as *B. amyloliquefacience* and *P. psychrotolerances* (data not shown). These results suggest that *RD29A* and *RD29B* are important regulators that fine-tune the parts of ABA- and drought-responsive signaling pathways in priming drought tolerance and promoting growth of soybean plants.

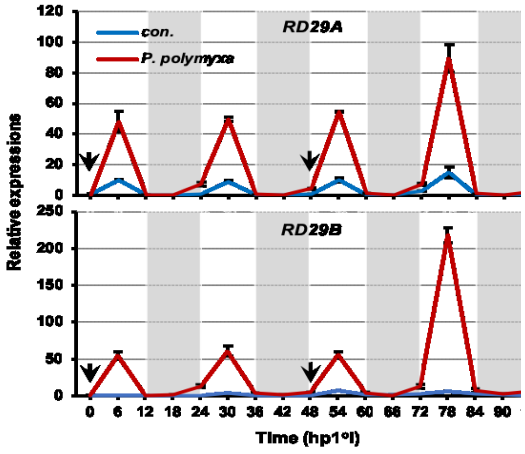


Figure 2. Circadian clock-dependent regulations of *P. polymyxa* mem- ory genes. Time-course real-time qRT-PCR of *RD29A* and *RD29B* in soybean, grown under 12-hr light/12-hr dark conditions, following the root inoculation of *P. polymyxa* (2x 10mL of 10^8 cfu•mL^{-1}). Total RNAs were prepared by every 6 hp1°i (starting from 9 am, arrow) for 96 hours. Note that the 2nd inoculation of *P. polymyxa* was carried out at 48 hp1°i (arrow), and values were normalized to the expression of *GAPDH* (means ± SD; n = 3). Grey boxes indicate the dark condition

3. *P. polymyxa* also upregulate the expression of *ABCG13*, a known drought memory gene. A previous study reported that *RD29B*, along with *LTI30*, *RAB18* and *ABCG13*, is - in fact - a drought memory gene (BMC Plant Biol 13: 229). However, *P. polymyxa* showed little effect on the expression of *LTI30* and *RAB18* mRNAs, but upregulation of *ABCG13* transcripts (Fig 3), describing that *P. polymyxa* activate only part of drought-responsive mechanisms that is perhaps independent from stomata closure processes, so that being able to prime drought tolerance without compromising plant growth and development.

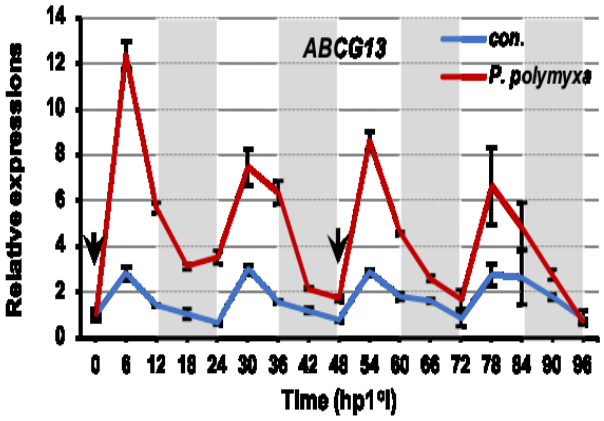


Figure 3. Upregulation of a drought memory gene, *ABCG13*, by *P. polymyxa*. Real-time qRT-PCR of *ABCG13* in soybean, grown under 12-hr light/12-hr dark conditions, following the root inoculation of *P. polymyxa* (2x 10mL of 10^8 cfu•mL^{-1}). Total RNAs were prepared by every 6 hp1°i (starting from 9 am, arrow) for 96 hours. Note that the 2nd inoculation of *P. polymyxa* was carried out at 48 hp1°i (arrow), and values were normalized to the expression of *GAPDH* (means ± SD; *n* =3). Grey boxes indicate the dark condition.

Future direction is to utilize reverse genetic (knock-out) approach in Arabidopsis and corroborate the phys- iological functions of *RD29A* and *RD29B* in plant growth and drought priming processes. These experiments will confirm the suitability of *RD29A* and *RD29B* in the use as transgenes to generate new transgenic drought tolerance lines in soybean and other plants.

Evaluation of Fungicide Treatments for Management of Soybean Rust on Soybean in South Alabama, 2018

W. Sanchez, k. S. Lawrence, W. Groover, D. Dyer, K. Gattoni, B. R. Lawaju, and M. N. Rondon

A fungicide trial was conducted to evaluate the efficacy of selected fungicide products on the soybean cultivar Asgrow AG 74X8 against soybean rust at the Gulf Coast Research and Extension Center in Fairhope, AL. The soil class was a Malbis sandy loam composed of 59% sand, 31% silt, and 10% clay. The previous crop was wheat. The field was strip-tilled and seed was sown on 19 Jun. Plots consisted of four rows that were 25 feet long with a 3.3 feet row spacing, arranged in a randomized complete block design with four replications and separated by a 6 ft fallow alley. Plots were maintained with standard herbicide, insecticide, and fertility production practices as recommended by the Alabama Cooperative Extension Service throughout the season. An overhead, lateral irrigation system was used to provide irrigation as needed. The five treatments included a non-treated, Aproach Prima, Priaxor and Stratego YLD applied at the R2 growth stage and Aproach Prima applied at R2 and R4. Fungicides were broadcast through Greenleaf technologies (turbo drop) TDXL 11002 spaced 30-in. apart on all four plot rows using a Lee Spider self-propelled sprayer. The sprayer was calibrated to deliver 15 gal/A at 50 psi. All treatments were evaluated at R6 for percent defoliation and percent soybean rust. Defoliation evaluations were made based on observations of the two center rows per plot and using a percent scale where 0%=no defoliation and 100%=complete defoliation of all of the plants within the two center rows. Soybean rust percentages were calculated using a 0-100% scale where 0%= no soybean rust pustules present on the abaxial side of the leaves, and 100%= disease present on all plants in the two center plot rows. Yield data (bu/A) were recorded at physiological maturity (R8). The center two rows of each plot were machine-harvested on 20 Oct using a small-plot combine equipped with an onboard weigh system and moisture meter. Plot weight was standardized to 13% moisture to calculate yield (bu/A). Data were analyzed using PRO GLIMMIX in SAS (Version 9.4, SAS Institute, Inc., Cary, NC) and means were compared to the non-treated control using the LSMEANS statement with the adjustment for Dunnett's method at $\alpha = 0.1, 0.05, 0.01$, and 0.001. Additionally, pairwise comparisons of treatment means were also made using the LSMEANs with the option for the Tukey HSD adjustment ($\alpha = 0.05$) to control for familywise error.

Monthly maximum temperatures from planting in June through harvest in October were 88.4, 89.5, 87.4, 88.0, and 82.8°F with average minimum temperatures of 73.3, 73.9, 72.1, 72.5, and 62.7°F, respectively. Rainfall accumulation for each month (Jun to Oct) was 4.6, 6.1, 9.2, 10.1, and 3.5-

in. respectively, with a total of 33.6-in. during the growing season. There was a significant effect of fungicide treatment on percent defoliation ($P = 0.005$). Not surprisingly, there was also significant effect of treatment on soybean rust severity ($P < 0.0001$). Percent defoliation and soybean rust severity were significantly lower ($P < 0.0001$) in all treatments as compared to the non-treated control, but there were no differences among treatments. Hence foliar applications were effective soybean rust management. Interestingly, there was a significant difference in yield between the Approach Prima applied at R2 and the Approach Prima applied at R2 and R4 compared to the non-treated at $\alpha = 0.1$.

		Percent evaluation (0 to 100%)		
Treatment, rate (fl oz/A)	Application Growth Stage[w]	Defoliation [x,y]	Soybean rust [z]	Yield (bu/A)
Non-treated		37.5 a	12.5 a	67.8
Aproach Prima 2.34 SC, 6.8 oz/A	R2	13.8 b***	1.5 b****	71.8*
Priaxor 4.17 SC, 8 oz/A	R2	13.8 b***	2.0 b****	70.4
Stratego 250 EC, 10 oz/A	R2	17.5 b***	2.5 b****	67.9
Aproach Prima 2.34 SC, 6.8 oz/A	R2, R4	10.0 b****	1.5 b****	71.9*

[z] Percent soybean rust was based on evaluating plants using a 0-100% scale where 0%= no disease present and 100%= disease present on all plants evaluated.
[y] Means in the same column followed by * ($\alpha = 0.1$), ** ($\alpha \leq 0.05$) ,*** ($\alpha = 0.01$) and **** ($\alpha = 0.001$) are significantly different from one another according to Dunnett's and Tukey's HSD.
[x] Defoliation percentage calculated using a 0 to 100% defoliated rating scale
[w] Plant stage at which the treatment was applied

Statewide Monitoring For Soybean Diseases, 2018

E. Sikora, D. Delaney, and K. Conner

The goal of this project was to continue the statewide soybean disease-monitoring program to alert farmers of early detection of yield-reducing foliar diseases, as well as of emerging pathogens that may threaten soybeans in the long-term. The monitoring program consists of biweekly scouting of soybean sentinel plots and in-field scouting of commercial fields by Extension Specialists and Regional Extension Agents. A primary focus is placed on identifying outbreaks of foliar diseases such as soybean rust (SBR), frogeye leaf spot (FLS), Cercospora leaf blight (CLB) or target spot that pose a significant threat to crop in-season, but we also need to learn more about emerging diseases such as taproot decline (TRD) and *soybean vein necrosis virus* (SVNV).

SBR was did not pose a significant threat for a significant portion of the state in 2018. The disease was found in only 13 of the 67 counties in Alabama (Figure 1). Many of these reports were from kudzu or maturing soybeans late in the growing season. We suspect a few stretches of freezing weather in the winter and early spring did not allow the pathogen to overwinter on kudzu in south Alabama or the Florida panhandle. This reduced the amount of disease inoculum in the region prior to planting. Relatively dry conditions in July and August in many parts of the state also inhibited disease development.

Significant outbreaks of FLS, CLB and target spot were observed at high levels in a limited number of fields in 2018. However, in most cases, disease pressure was not at levels that would cause significant yield loss. As if often the case, high levels of CLB were more concentrated in fields in southwest Alabama. Yield losses were apparent on some late maturing cultivars in an experimental block at the AAES Research Station in Fairhope. Fungicides for the most part are not that effective in reducing damage from CLB.

Incidence of a newly emerging disease (TRD) continued to increase in Alabama. The pathogen was detected in six new counties in the state in 2018 including Baldwin, Clarke, Colbert, Covington, Lauderdale, Lawrence and Mobile (Figure 2). This brings the total number of counties in Alabama reporting the disease to 22 since 2016. In working closely with plant pathologists at Louisiana State University, we have been able to determine that the genetic variation in populations of TRD in the southeast is not very high, suggesting that developing a rapid test to determine disease resistance among soybean cultivars may be possible. TRD is a developing problem for soybean growers in the south, and researchers are just beginning to understand its importance and epidemiology.

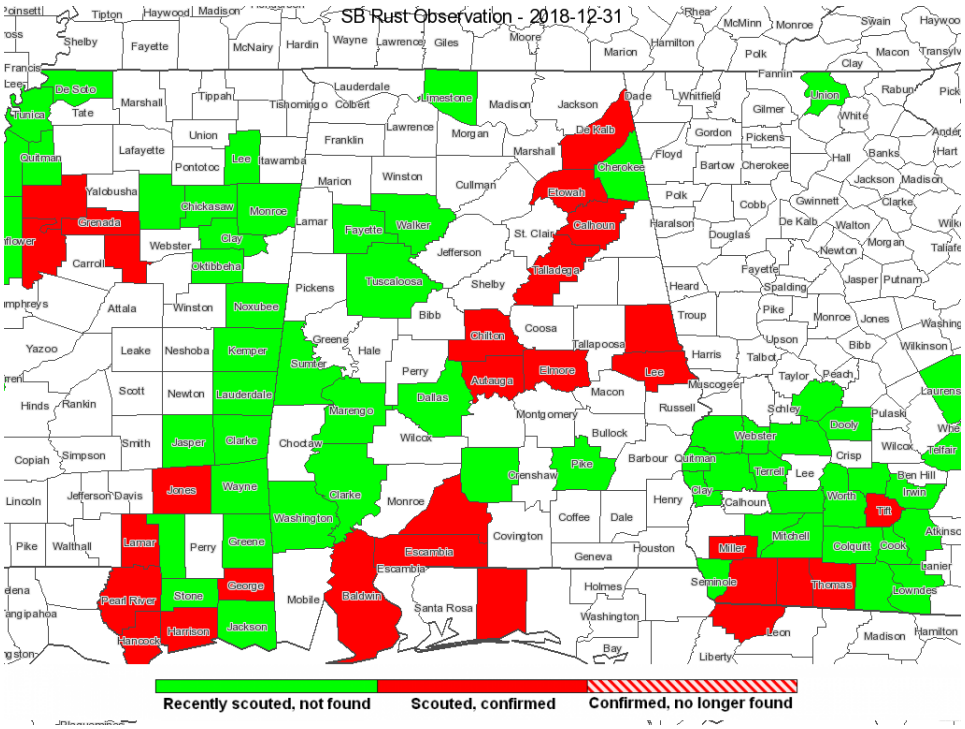


Figure 1. Soybean rust distribution in Alabama and the Southeast in 2018.

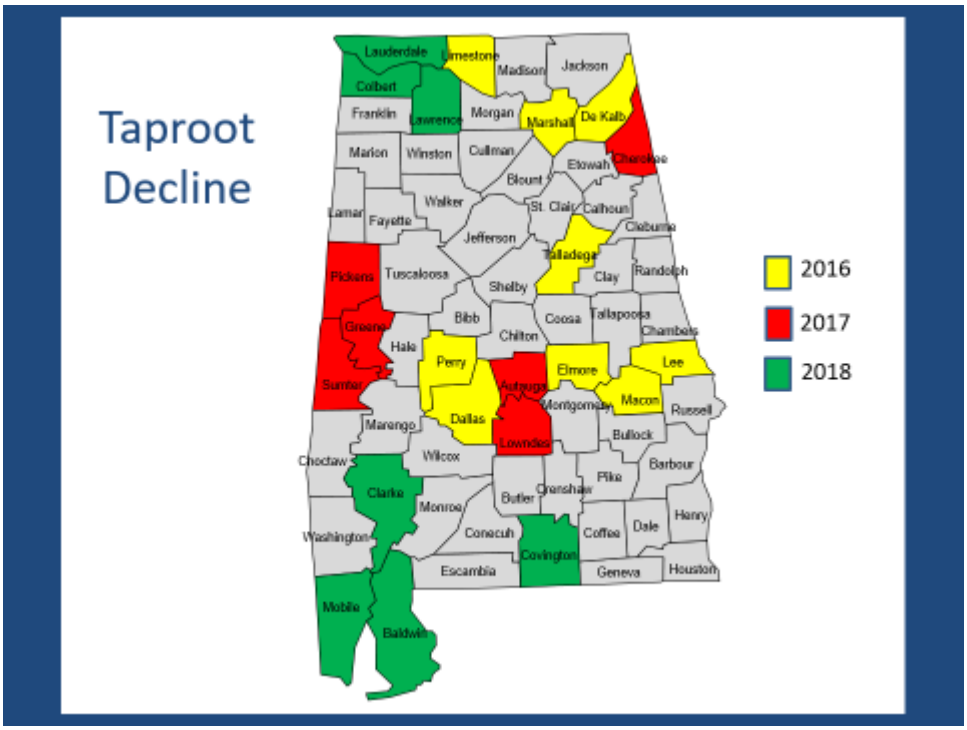


Figure 2. Counties reporting taproot decline in 2016 (yellow) and 2017 (red) and green (2018).

V. Insect Management

Impact of the Parasitic Wasp as Biological Control for Suppressing Kudzu Bug Populations in Soybeans

X. P. Hu and T. Reed

In Alabama, the exotic kudzu bug was discovered in 2 counties in 2010. It quickly made its way across the 67 counties by August 14, 2013. At the same time, this insect became a key pest of soybean crops. Significant yield loss up to 60% was in field if left unchecked in GA and up to 40% yield loss in AL if not repeatedly sprayed, reported by Tim Reed. The loss in yield is attributed to plant stress from its feeding activities.

With little information regarding this exotic insect, insecticide spray was recommended as the only method to control this pests. Economic threshold was recommended as 2 adults per sweep (5-ch diameter net) on field edges (Soybean Grower). This resulted in multiple sprays throughout the field season from vegetative to maturation stage and excess costs.

As we learned more about its biology (life cycle, aggregated distribution at field edges), another threshold of 1 nymph per sweep was recommended. One sweep consists of a side-to-side arc of the net 180º through the soybean canopy. A single well-timed insecticide application at soybean R2 – R3 growth stage targeting nymphs was reported as effective as multiple sprays (Seiter et al. 2015).

The discovery of natural enemies in 2013 could be the change point of control game. We made three discoveries in soybean fields on Auburn: a non-native braconid egg parasitoid wasp that specifically attacks kudzu bug, a native parasitoid fly that attacks kudzu bug adults, and an indigenous entomopathogenic fungus, *Beauveria bassiana*, that infects kudzu bug nymphs and adults. Of the 3, the most interesting is the exotic egg parasitoid wasp, *Paratelenomus saccbraralis*. Literatures record it the most effective parasitoid holding kudzu bug at bay in Asia. It is still uncertain how the Asian wasp made it to the US, likely having arrived accidentally via trade, just as the kudzu bug did.

We conducted this 7-year project from 2012-2018. Our short-term goal is to understand if insecticide spray would harm both the pest and natural enemy and if the egg parasitoid occurrence is limited locally. Our mid-term goal is to gain knowledge about relationships among insecticide, kudzu bug, and egg parasitoids, and help soybean growers make control decision accordingly. Our ultimate goal is to develop cost-effective and agro-ecosystem friendly kudzu bug management program. The specific objectives are:

Objective 1: Investigate the effect of insecticides on populations of soybean kudzu bugs and egg parasitoids.

Objective 2: Evaluate the impact of egg parasitoid wasp on suppressing soybean kudzu bug.

Objective 3: Monitor the range expansion of the egg parasitoid in AL.

Methods used in this project

Systematic surveying technology was employed for objective 1 and 2. The surveys were conducted in soybean growth season at three research centers (Prattville, Brewton) and Auburn campus. There were 4 soybean fields at Prattville and Brewton (each had 3 sprayed and 1 unsprayed), and 1 unsprayed field on AU campus. Kudzu bug populations were estimated by counting 4 samples of 5 plants from each field; egg parasitism rates were estimated by examining 10 egg-masses per field. Data were collected biweekly.

Random sampling technique was used for objective 3. Samplings were collected from soybean fields or kudzu patches throughout Alabama. At least 20 egg-mass were examined per visit to each county. Parasitized eggs were identified by color difference or bringing back suspicious egg-masses to laboratory to observe wasp-emerging.

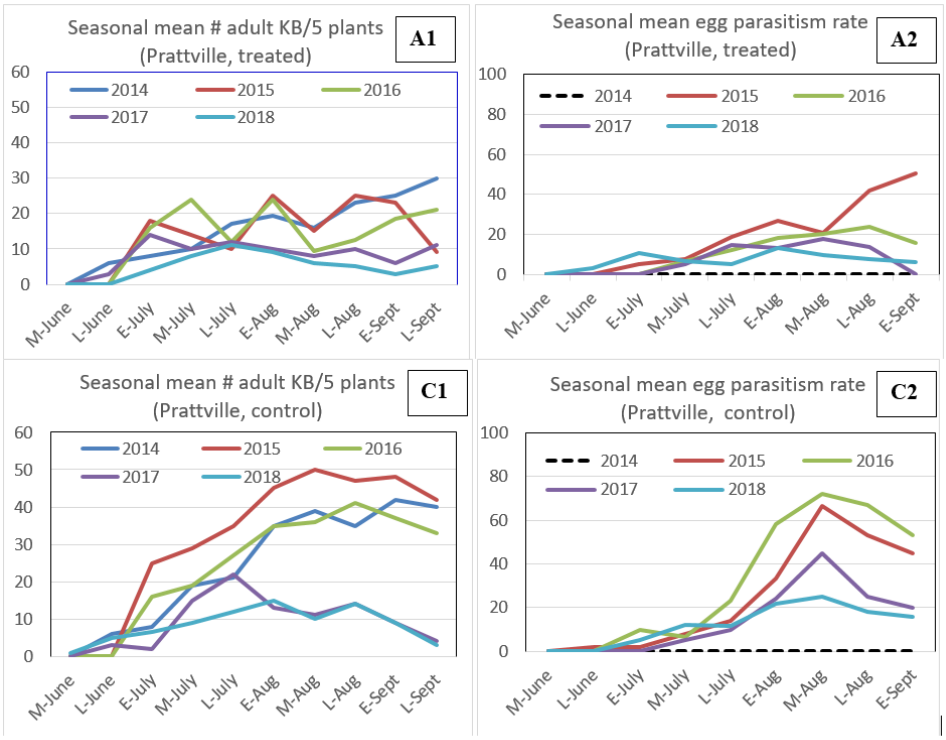
Results

Objective 1: The effect of insecticides on populations of soybean kudzu bugs and egg parasitoid.

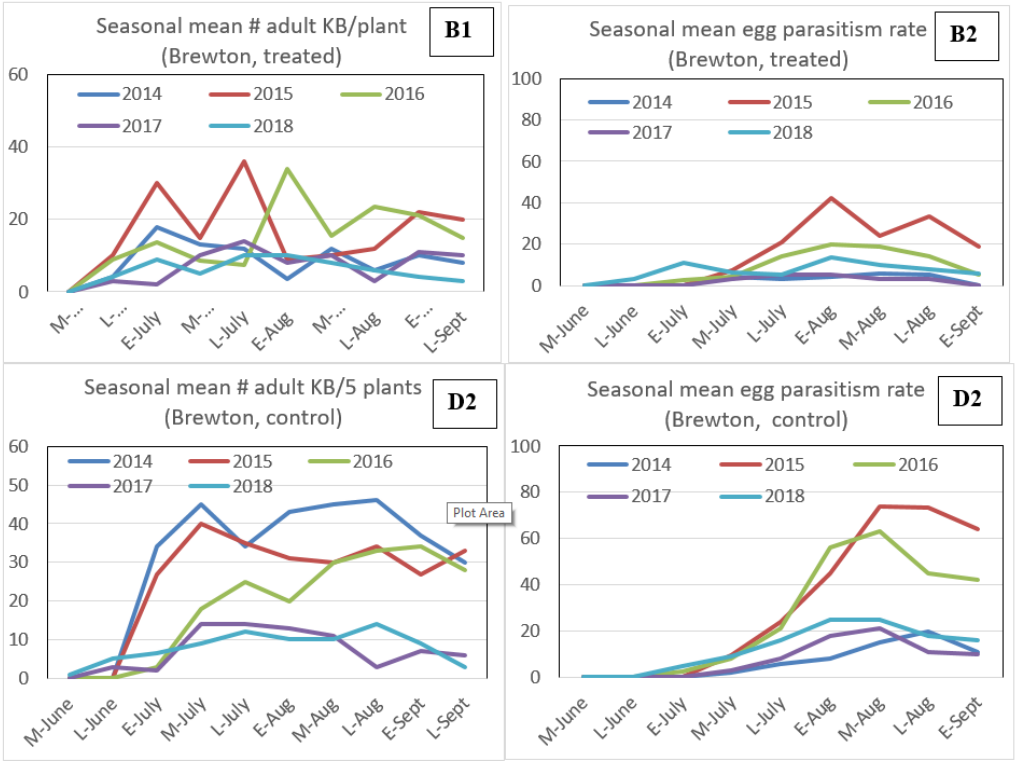
At Prattville, data from sprayed fields indicate that insecticide sprays were effective in killing kudzu bugs but also suppressed egg parasitism rates: KB populations were lower in treated field (Fig A1) than control (Fig C1); egg parasitism rates were lower in treated (Fig. A2) than in control (C2).

Insecticide sprays kept kudzu bug density below 30 adults/ 5-plants in 2014, 2015, and 2016, and below 10 adults/5-plants in 2017 and 2018.

Kudzu bug populations were higher in 2014, 2015 and 2016; population declined in 2017 and 2018.

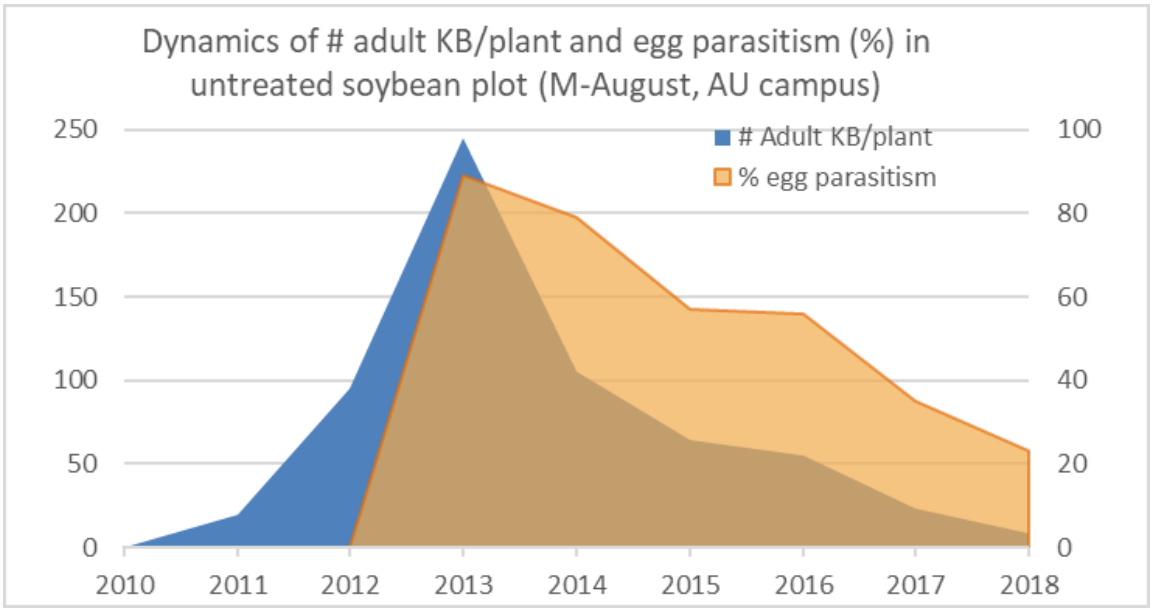


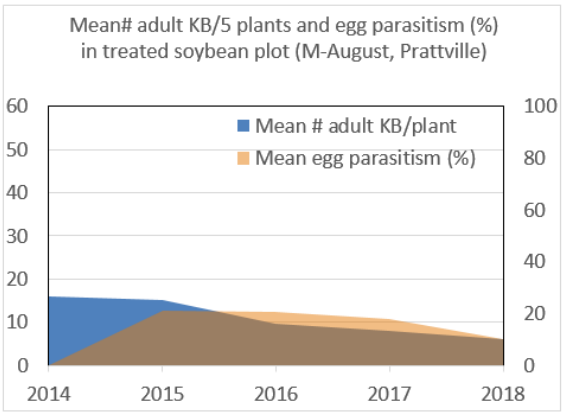
At Brewton, we saw similar result and trend. Insecticide prays sprays suppressed both kudzu bugs (Fig B1 vs. Fig B1) but also suppressed egg parasitism rates (Fig. D2 vs. Fig D2).

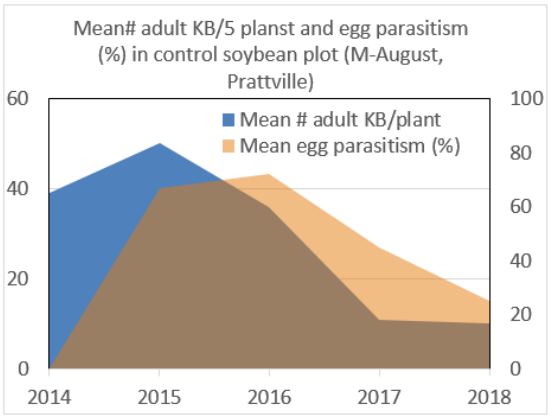


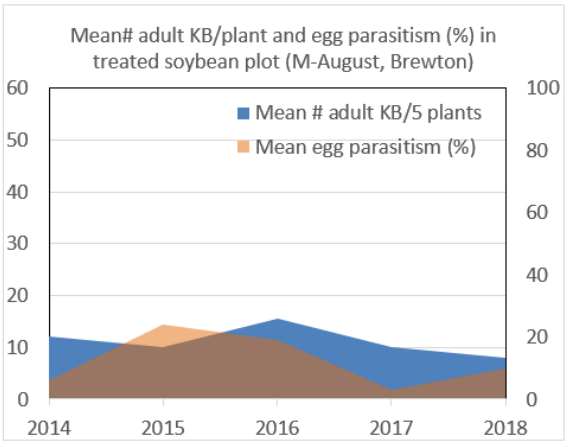
Objective 2: Impact of egg parasitoid wasp on suppressing soybean kudzu bug.

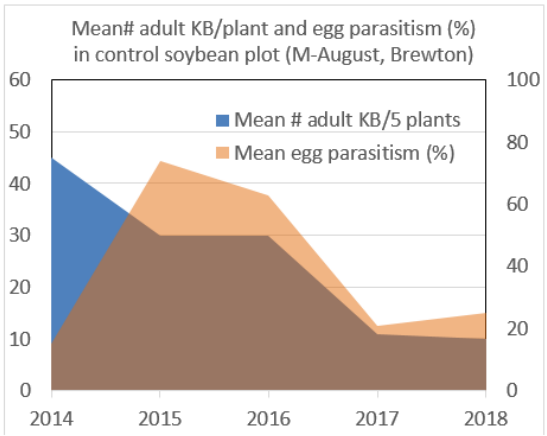
Data collected in Mid-August are present in the following figures.







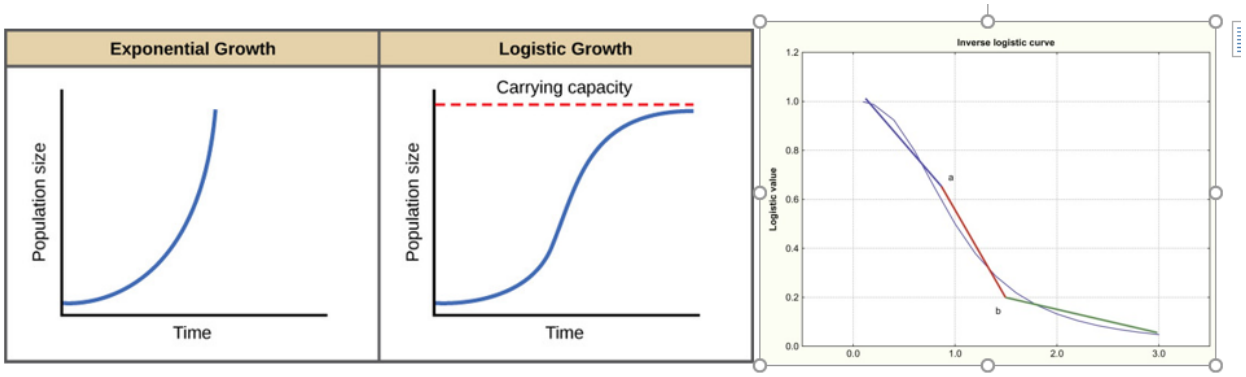




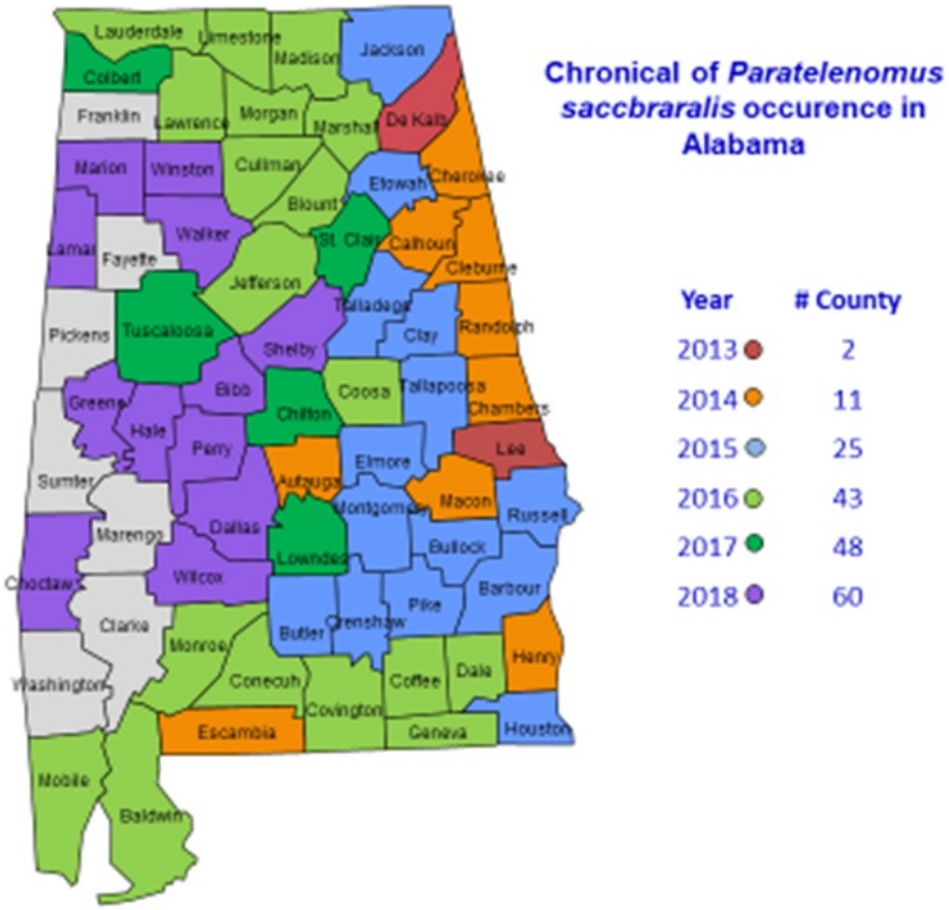
The dynamics of kudzu bug density and wasp parasitism rate collected in Mid-August from unsprayed soybean fields shows:

- If left unsprayed, kudzu bug population had an **Exponential Growth** during the first 3 years when the egg parasitoid was absent, then level off shortly (kind of a **logistic growth**),

the decline from year 4 through year 7, as the function of the parasitoid wasp (**inverted logistic growth**).



Objective 3: Range expansion of the egg parasitoid in AL.



Egg parasitoid wasp continue disperse across AL. It has been confirmed in 60 out of the 67 Counties in Alabama. Of the 7 unconfirmed counties, 2 were visited but did not find parasitoid wasp, the other 5 have not been surveyed yet.

- Insecticide spray kill both kudzu bug and egg parasitoid. To minimize the negative effect on egg parasitoid, we recommend the spray should be timed before egg-hatching peak period that is usually at soybean R1-R2 growth stage.
- With absence of egg parasitoid wasp, if left unchecked, kudzu bug populations are able to increase exponentially during the first couple of years, then a logistic growth pattern. Insecticide spray should be advised.
- However, when egg parasitoid is present, insecticide spray may do little to suppress kudzu bug populations. Our data show similar kudzu bug populations regardless of spray or not, indicating that the wasp is capable for keeping kudzu bugs at bay. If a spray is necessary, it should be a well-timed to avoid harm the wasp. Based on our preliminary data, we propose a threshold of 10 adults per 5 soybean plants. Future study is needed to determine a threshold considering natural enemy factors.
- The wasp has dispersed to 60 Counties, and is likely to spread across AL.
- Kudzu bug numbers have been dwindling since 2013, so do the egg parasitoid wasp. We need to closely monitor their populations and prepare for possible resurgence of the pest.

Efficacy of Bt Soybeans in Preventing Yield Loss to Soybeans

T. Reed and R. Smith

Materials and Methods

A total of 27 different entries were planted on June 28, 2018 at the Gulf Coast Research and Extension Center at Fairhope. The entries were all derived from one Group 3.5 soybean variety and no entry possessed a GMO herbicide-resistant trait. One entry lacked a Bt protein while all the other experimental lines possessed novel Cry proteins.. The study also included the Intacta and Intacta 2 soybeans. There were 4 replications for each entry with entries arranged in a RCB design. Each plot was 4 rows wide and 30 feet long with a row spacing of 38 inches. A 4 row buffer of Bt soybeans was planted between each plot and along the outer perimeter of the trial. Soybeans were grown using standard production practices. Amdro was applied to the test area on August 13 to reduce fire ant numbers. Plots were sprayed with 2 oz/acre of Centric to reduce beneficial arthropods on August 13 also. Plots were sampled for caterpillars by vigorously shaking plants over a 3-foot wide ground-cloth in each row of each plot on each sampling date. Plots were sampled 7 times beginning on 7/24 and ending on 9/20. Percent defoliation was estimated on each sampling date. Yields were not taken in this study.

RESULTS: .SBL's were first found in the trial in non-Bt soybeans on 8/15 when soybeans were in the R 3.5 stage of development. By August 23 numbers of SBL's in the non-Bt plots had increased from 18 /3 row ft to a peak number of 60.5/3-row ft. GMO entry #6 had 11.5 SBL/3 row ft and 1% defoliation on 8/23. Entry # 7 had 1.25 SBL/3-row ft. Entries 9 and 10 also had less than 1 SBL/3-row ft. % Defoliation reached 31% by 8/31 in the non-Bt plots which averaged 35 SBL/3-row ft. Entry 6 had 8 SBL/3-row ft and 2% defoliation. Defoliation increased slightly to 34% by September 7 as the SBL numbers in the non-Bt entry dropped to 6/3-row ft. Velvetbean caterpillar larvae were detected for the first time in the trial on 9/07 with Entries 7 and 21 having mean densities of 7.5 and 3.75/3-row ft. respectively with both Entries having less than 1% defoliation. The non-Bt entry had 0.5 VBC/3-row ft . VBC numbers jumped to 21/3-row ft in non-Bt plots by 9/20. Densities of VBC larvae in Entries 7 and 19 reached 15 and 3 VBC/3-row ft with corresponding defoliation levels of 11% and 3% respectively. Entry 3 had 1 VBC/3 row ft and 0.75% defoliation. The presence of both SBL and VBC larvae in this study proved quite useful in assessing efficacy of the entries in preventing feeding by both species. Entries 6 and 7 were fed upon by SBL. Entries 7, 19 and 21 were fed upon by VBC. All the other experimental lines did not incur feeding damage from either caterpillar .Both the Intacta and the Intacta 2 traits performed very well in the study.

Table 1. Mean number of soybean looper and velvetbean caterpillar larvae per 3 row feet and percent defoliation for Bt soybean entries on 5 different dates at Fairhope, AL in 2018.

	8/15			8/23			8/31			9/07			9/11			9/20		
Entry	**SBL[1]**	**VBC[2]**	**%Def**	**SBL**	**VBC**	**%Def**	**SBL**	**VBC**	**%Def**	**SBL**	**VBC**	**%Def**	**SBL**	**VBC**	**%Def**	**SBL**	**VBC**	**%Def**
1	18	0	3	60.5	0	9.5	35.0	0	31.0	6.25	0.5	34.0	2.75	23.75	44.0	0	21	50
2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0.75
4	0	0	0	0	0	0	0	0	0	0	0	0	0	0.3	0	0	0	0.5
5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
6	0	0	0	11.5	0	0.75	8.0	0	2.0	0.75	0	0.5	0	0	0	0	0.3	2.5
7	0	0	0	1.25	0	0	0	0	0	0	7.5	0.25	0	0	0	0	15	11
8	0	0	0	0	0	0	0.25	0	0	0	0	0	0	0	0	0	0	0.5
9	0	0	0	0.25	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
10	0	0	0	0.75	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
11	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.3	0.75
12	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.75
16	0	0	0	0.25	0	0	0.25	0	0	0	0	0	0	0	0	0	0	0.5
17	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
18	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
19	0	0	0	0.25	0	0	0	0	0	0	0	0	0	0	0	0	3	3.25
20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.75
21	0	0	0	0	0	0	0	0	0	0	3.75	0.25	0	0	0	0	0	0.5
22	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
23	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
24	0	0	0	0	0	0	0	0	0.25	0	0	0	0	0	0	0	0	0.5
25	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
26	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5
27	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.5

Developing Optimal Management Strategies for Key Insect Pests of Soybeans

T. Reed

Objectives 1 and 2

Comparative Insecticide Efficacy In Controlling Soybean Loopers Infesting Soybeans When Insecticides Are Applied Pre- and At- Threshold

This study was conducted at the Brewton Agricultural Research Unit. Soybeans were planted using a 36 inch row spacing on 6/6. The variety P47T89R was planted in the Pre-Threshold SprayTest and the variety P55T81R was planted in the At-Threshold Spray test.Plot size was 8 rows wide x 27 ft long. The Insecticide Treatments for the Pre-Threshold spray test are presented in Table 1. The At-threshold Spray test included the pyrethroid Delta Gold instead of Orthene. Pre-Threshold sprays were applied on August 3. Pre-threshold spray counts for soybean loopers (SBL's) = 0.5 large (L) +8 medium (M) + 0.9 small (S) larvae per row ft. Pre-Threshold spray counts for green clover worms (GCW.s) = 0.1 L + 0.23 M + 1.1 L per row ft. At-Threshold sprays were applied August 10. Pre-spray sweep net counts Aug 9 = 1.0 SBL/sweep (0.4 L + 0.25 M + 0.3 S) and 1% defoliation. Chemicals for both the Pre and At-threshold tests were applied in 10 gallons of water/acre using J.D. PSLADQ 10015 nozzles and 40 psi. After sprays were applied larval populations were sampled by taking 10 sweep net samples across two rows in each plot on each of 3 sampling dates (8/15, 8/24, 8/29). Drop cloth samples were taken from 6 row feet in each plot on the last sampling date (9/11 Pre and 9/12 At-Threshold).).

Results: Numbers of soybean loopers recovered in sweepnet samples at 12, 21 and 26 days after the insecticides were applied (DAA) are presented in Table 1. Numbers of SBL's and velvetbean caterpillars (VBC,s) recovered in drop-cloth samples at 39 DAA are presented in Table 2.

Table 1. Total Number of Soybean Loopers Per 10 Sweeps and Per Cent Defoliation at 12, 21 and 26 DAA When Sprayed Pre-Threshold – Brewton AL 2018

		8/15 – R 5.4		8/24 – R 5.7		8/29 – R 6.0	
Treatment	Rate per acre	SBL/10SW	% DEF	SBL/10SW	% DEF	SBL/10SW	% DEF
Baythroid XL	2.8 oz	10.8 a	1.8 ab	46.3 ab	4.8 a	15.0 abc	7.0 a
Mustang Max	4.0 oz	10.5 a	1.5 abc	52.5 a	5.0 a	17.5 ab	5.6 ab
Ammo	4.0 oz	11.5 a	1.0 c	36.3 c	4.5 ab	12.5 bcde	6.0 ab
Warrior II Z	1.92 oz	9.3 a	2.0 a	39.3 bc	5.3 a	14.5 abcd	4.3 bc
Bifenthrin	6.4 oz	10.0 a	1.0 c	39.3 bc	3.5 bc	19.8 a	3.0 cd
Prevathon	14.0 oz	2.8 c	1.3 bc	23.5 e	1.0 e	8.0 e	2.3 d
Besiege	8.0 oz	3.3 c	1.0 c	24.3 de	1.3 e	8.5 de	1.8 d
Dimilin	4.0 oz	4.8 bc	1.3 bc	33.3 cd	3.0 cd	14.5 ab	4.5 bc
Orthene 97	1.0 lb.	4.0 c	1.5 abc	42.0 bc	2.0 de	17.0 ab	4.0 bc
Unsprayed	---	8.3 ab	2.0 a	36.0 c	4.8 a	10.0 cde	5.5 ab
	X% small larvae =	39.7 %		62.6 %		42.3 %	
	P > F =	0.0014	0.0058	0.0004	0.0016	0.04	0.0000
	LSD 0.1 =	4.0	0.51	9.4	1.25	6.2	1.14

Numbers of SBL's ranged from 2.8 in the Prevathon Treatment to 11.5/10 sweeps in the Ammo treatment at 12 DAA when about 40% of the larvae were small. Defoliation ranged from 1% to 2%. Larval populations peaked at 21 DAA and 63% of the larvae were small. SBL numbers per 10 sweeps ranged from23.5/10 sweeps in the Prevathon Treatment to 52.5 with Mustang Max. Defoliation ranged from 1% to 5%. SBL numbers were declining by 26 DAA and the untreated plots had 10 SBL/10 sweeps, 72% less than 5 days earlier. About 40% of the larvae were small. Defoliation changed very little between 21 and 26 DAA Defoliation is lowest in the Prevathon and Besiege Treatments. Defoliation in the bifenthrin Treatment is noticeably lower than in 3 of the other pyrethroid Treatments.

Table 2. Total Number of SBL's Per 6 Row Ft and Per Cent Defoliation at 39 DAA When Sprayed Pre-threshold-Brewton, AL 2018.

Treatment	Rate per acre	SBL/6 row ft L+M	SM	VBC/6 row ft L+M	SM	% DEF	
Baythroid XL	2.8 oz	1.3	2.0	1.8	6.0	17.0	AB
Mustang Max	4.0 oz	2.8	1.5	1.3	5.5	15.0	BC
Ammo	4.0 oz	3.3	2.3	2.3	3.3	16.8	AB
Warrior II Z	1.92 oz	0.0	1.8	0	6.5	16.3	ABC
Bifenthrin	6.4 oz	1.0	2.8	0	0.0	12.0	CD
Prevathon	14.0 oz	0.8	0.3	0	0.0	2.0	E
Besiege	8.0 oz	0.8	1.8	0	0.3	2.0	E
Dimilin	4.0 oz	0.8	2.0	0.8	0.8	10.0	D
Orthene 97	1.0 lb	0.8	3.3	2.8	5.5	10.0	D
Unsprayed	---	1.5	2.8	4.5	13.5	20.0	A
	P>F =	0.008	0.870	0.05	0.03	0.0000	
	LSD 0.1 =	1.3	---	2.4	6.4	4.3	

The number of large SBL's per 6 row ft (1.5) at 39 DAA was less than that for velvetbean caterpillar (VBC) larvae (4.5). VBC's accounted for much of the increase in defoliation between

26 and 39 DAA. The defoliation level in untreated plots was not significantly different from that in the Baythroid XL, Ammo and Warrior II Z Treatments. Bifemthrin was the only pyrethroid in which no VBC larvae were recovered at 39 DAA.

At-Threshold Spray Results: Numbers of soybean loopers recovered per 10 sweepnet samples at 5,13 and 19 DAA are presented in Table 3. SBL numbers and per cent defoliation was similar to that in the Pre-threshold spray test. Although there was no significant difference in the number of SBL larvae present per 10 sweeps, the defoliation level in the bifenthrin treatment at 19 DAA was significantly less than that in the other 5 pyrethroid sprays .

Table 3. Total Number of Soybean Loopers Per 10 Sweeps and Per Cent Defoliation at 5, 13 and 19 DAA When Sprayed At-Threshold – Brewton AL 2018

		8/15		8/23		8/29	
Treatment	Rate per acre	SBL/10SW	% DEF	SBL/10SW	% DEF	SBL/10SW	% DEF
Baythroid XL	2.8 oz	12.5 bcd	2.8 ab	46.3 ab	7.0 a	19.0 ab	6.0 bc
Mustang Max	4.0 oz	19.5 a	4.3 a	52.5 a	5.8 ab	17.0 abc	6.3 b
Ammo	4.0 oz	19.5 a	3.8 ab	36.3 c	6.0 ab	17.8 abc	7.3 ab
Warrior II Z	1.92 oz	15.5 abc	3.0 bc	39.3 bc	4.3 bc	15.3 bc	7.3 ab
Bifenthrin	6.4 oz	9.5 def	2.5 cd	39.3 bc	3.0 cd	19.0 ab	4.5 c
Prevathon	14.0 oz	6.0 ef	2.0 d	23.5 e	1.5 d	11.8 cd	2.3 d
Besiege	8.0 oz	5.0 f	2.0 d	24.3 de	1.8 d	8.0d	2.0 d
Dimilin	4.0 oz	10.5 cde	3.3 bc	33.3 cd	4.5 bc	16.5 abc	5.8 bc
Delta Gold	2.4 oz	16.0 ab	3.5 ab	42.0 bc	4.0 bc	23.3 a	8.8 a
Unsprayed	---	11.8 bcd	3.3 bc	36 c	5.5 ab	13.8 bcd	7.3 ab
	X% small larvae =	38.3 %		80 %		60 %	
	P>F =	0.0003	0.0021	0.0004	0.0016	0.077	0.000
	LSD 0.1=	2.2	0.9	9.4	2.1	7.2	1.65

Numbers of SBL's and velvetbean caterpillars (VBC"s) recovered in drop-cloth samples at 32 DAA are presented in Table 4.

Table 4. Total Number of SBL's and VBC's Per 6 Row Ft and Per Cent Defoliation at 32 DAA When Sprayed At-threshold-Brewton, AL 2018.

Treatment	Rate per acre	SBL/6 row ft		VBC/6 row ft		%	
		L+M	SM	L+M	SM	DEF	
Boythroid XL	2.8 oz	3.0	1.0	1.0	6.8	23.8 a	
Mustang Max	4.0 oz	2.0	1.8	3.0	7.0	25.0 a	
Ammo	4.0 oz	1.8	1.8	1.8	9.0	25.0 a	
Warrior II Z	1.92 oz	2.8	3.3	2.8	7.8	21.3 a	
Bifenthrin	6.4 oz	1.5	3.3	0	2.0	13.8 b	
Prevathon	14.0 oz	1.5	1.3	0	0.5	4.0 c	
Besiege	8.0 oz	0.8	1.5	0	1.3	4.0 c	
Dimilin	4.0 oz	2.0	1.3	0.3	1.0	23.8 a	
Delta Gold	2.4 oz	1.0	1.5	1.3	12.3	26.3 a	
Unsprayed	---	1.5	3.4	1.5	10.5	26.3 a	
	P>F =	0.71	0.35	0.038	0.007	0.0000	
	LSD 0.1 =	---	---	2.02	2.21	5.76	

Bifenthrin gave the best control of VBC's among pyrethroids in the At-threshold spray test. Comparison of defoliation levels on 9/11-12 for the Pre- and At-threshold spray tests show a consistent trend toward higher defoliation levels for all treatments in the At-threshold spray test. The Dimilin treatment showed the greatest numerical increase in per cent defoliation with a delayed application.

Objective 3

Determine the effect of pyrethroid insecticides on the stink bug complex infesting soybeans.

This study was conducted at the Brewton Agricultural Research Unit. P48T27X soybeans were planted using a 36 inch row spacing on 4/28. Plot size was 8 rows wide X 30 ft long. Insecticide treatments are presented in Table 1. Treatments included 6 pyrethroids, one organo-phosphate and one neonicotinoid insecticide with all applied at the maximum labeled rate. Insecticides were applied on 8/8 in 7 gallons of water/acre using TXVK6 Conejet nozzles and 45 PSI. Stink bugs counts were made 4 DAA by making 10 sweeps across 2 rows in each plot. No yields were taken in this trial.

Results: Results are presented in Table 1.

Table 1. Numbers of Stink Bugs and Kudzu Bugs Recovered at 4 DAA in 10 Sweepnet Samples Following Insecticide Applications to Soybeans . Prattville AL 2018

Treatment	Rate per acre	BMSB ad 10 sw	BMSB im 10 sw	SGSB ad 10 sw	SGSB im 10 sw	KB ad 10 sw	KB im 10 sw
Delta Gold	2.4 oz	0	0.5	0	0	2.3	0.8
Bifenthrin	6.4 oz	0	0	0.3	0	0.3	0
Belay	4.0 oz	0	0	0	0	0.8	12.3
Orthene	0.75 oz	0.5	0.3	0	0	1.5	0.8
Ammo	5.0 oz	0	0	0	0	0	0.8
Baythroid	2.8 oz	0.11	0.5	0	0.15	2.8	4.3
Mustang Max	4.0 oz	0	0.8	0	0	0.8	0.8
Warrior II	1.92 oz	0	0.3	0	0	0.3	0
Untreated	---	1.8	9.5	1.8	2.5	6.5	23.3
	P > F =	0.004	0.001	0.0073	0.01	0.095	0.21
	LSD 0.1 =	0.71	2.8	0.43	1.1	3.5	15.8

All insecticide treatments provided outstanding control of both Brown Marmorated stink bugs (BMSB) and Southern Green stink bugs. BMSB's comprised 50% and 79% of the adult and immature stink bug population, respectively.

Objective 4

Detemine The Impact On Yield When The Brown Marmorated Stink Bug is An Important Member of The Stink Bug Complex Feeding On Soybeans?

Materials and Methods: This study was conducted at the Prattville ARU. Plots were planted 4/28 using the variety P48T27X. Each plot was 8 rows wide and 30 feet long. Treatments were arranged in a RCB design with 8 replications of Treatment 1 and 16 replications of Treatment 2.. Treatments were as follows: Treatment 1-No Orthene applications; Treatment 2-Orthene 97 applied 7/17 and 8/6. The Orthene was applied at a rate of 1 lb/acre each application using TXVK6 Conejet nozzles, 45 psi and a 7 gallon spray volume/acre. Plots were sampled for pest insects on 4 dates using a 15 inch diameter sweep net. The 2 center rows of each plot were harvested 10/15 and yields were converted to 13% moisture. Results: Numbers of stink bugs recovered per 15 sweeps on 4 sampling dates are presented in Table 1.

Results: Numbers of stink bugs recovered per 15 sweeps on 4 sampling dates are presented in Table 1. BMSB's were the only stink bugs present on 7/12 when the stink bug population reached

the economic threshold. The first Orthene spray on 7/16 reduced stink bug numbers by 89% at 4 DAA. BMSB's accounted for 71% of the stink bugs present on 7/20. By 7/20 Southern green stink bugs had moved into plots and comprised about 30% of the stink bug population. The stink bug population rebounded and 21 days later stink bug numbers were similar in the treated and untreated plots. On 8/6 BMSB's comprised 66% of the stink bug population in unsprayed plots. Soybeans were in the R5.8 stage of development on 8/6 and were still susceptible to yield loss by stink bugs.The second orthene spray reduced the population by 91% .The BMSB comprised 66% of the stink bug complex on 8/20.

Table 1. Number of Stink Bugs per 15 sweeps on indicated sampling date.
Prattville AL 2018

	7/12 -R5.1		7/20 -R5.5		8/06 -R5.8		8/12 -R6.5	
Stink Bug	TRT 1	TRT 2	TRT 1	TRT 2	TRT 1	TRT 2	TRT 1	TRT 2
BMSB ad	3.0	2.4	6.6	0.9	6.0	5.85	3.45	0.45
BMSB im	0.15	0	0.45	0	11.1	5.85	15.9	0.75
SGSB ad	0	0	0.6	0	4.2	7.35	3.75	1.2
SGSB im	0	0	2.1	0.15	3.75	1.95	3.9	0.15
BSB ad	0	0	0.15	0	0.75	0.45	0	0
BSB im	0	0	0	0	0	0.15	0.15	0
SBT ot	3.15	2.4	9.9	1.1	25.8	21.6	27.15	2.55
% BMSB	100	100	71	82	66	54	71	47

TRT 1 had no insecticide. TRT 2 sprayed with one lb Orthene 97 on 7/17 & 8/6

There was a significant treatment effect with respect to yield (P>F = 0.0184). The 35.3 bu/ac in unsprayed plots were significantly less (LSD 0.1= 5.84) than the 44.2 bu/ac harvested in plots sprayed twice. This difference of 8.9 bu/ac at $10/bu represnts a loss of $89/acre at $10/bu .

Objective 5

Survey Alabama counties to determine the range of the brown marmorated stink bug (BMSB)

Materials and Methods:When this survey was initiated during the 2018 growing season the BMSB had already been found in 29 Alabama counties. During the survey a total of 16 counties were surveyed that had not been confirmed as having the BMSB. Soybean and cotton fields were sampled with sweepnets for this stink bug. Counties inspected were Baldwin, Conecuh, Covington, Dallas, Fayette, Geneva, Greene, Lauderdale, Marengo, Marion, Mobile, Monroe, Tuscaloosa, and Walker and Washington. Results: BMSB's were found in Lauderdale county in a soybean field on County Road 2 west of Florence. A total of 40 sweepnet sweeps netted two male BMSB's.

BMSB's were also collected in Walker county in the Nauvoo community on the side of a house. Some of the soybean fields that were inspected had been sprayed with insecticides and this reduced the chances of finding the stink bug. All cotton fields inspected had been sprayed previously. Surprisingly no BMSB's were found in the Blackbelt along Highways 80 and 5. A state map showing the current distribution of the BMSB is presented below.

Objective 6

Determine the effect of insecticide seed treatment, tillage, and foliar insecticide application on 3-cornered-alfalfa hopper (3CAH) girdling damage to soybeans.

Materials and Methods: This study was conducted at the Tennessee Valley Research and Extension Center at Belle Mina. The study was conducted using the variety P96Y70. Specific treatment combinations are presented in Table 1. There were 4 replications of each treatment combination arranged in a RCB design. Plots were 8 rows wide and 25 feet long. Bifenthin was applied to plots receiving a foliar spray on July 3 in 8.6 gallons of water per acre, 8X Conejet nozzles and 65 psi. Stand counts were taken July 6 and the number of plants girdled in each plot was determined on 8/17 by pushing over plants in 3 ft sections of row with a yardstick and examining the base of plants for girdles.

Results: Results are presented in Table 1. There was no significant treatment effect with respect to stand count (P>F = 0.94), % girdled plants (0.76) or yield (0.26). Plant stand counts ranged from

59 to 64 plants/6 row ft. Per cent girdled plants ranged from 4.7 to 7.4%. Yields ranged from 46.8 to 53.5 bushels/acre.

Table 1. Effect of Tillage Practice, Foliar Insecticide and Insecticide Seed Treatments on Stand Count, 3-Cornered Alfalfa Hopper Girdled Plants and Yield. Belle Mina, AL 2018.

Trt #	Tillage System	Insecticide Seed Trt	Foliar Insecticide	Plant/6 row ft	% Girdled Plants	Bu/Ac
1	No-Till Wheat Stubble	Yes	No	59	7.4	48.4
2	No-Till Wheat Stubble	No	No	61	7.3	53.5
3	No-Till Wheat Stubble	Yes	Yes	62	6.6	47.7
4	No-Till Wheat Stubble	No	Yes	62	4.7	48.8
5	Conventional Tillage	Yes	No	62	4.9	49.1
6	Conventional Tillage	No	No	64	7.3	46.8
			P>F =	0.94	0.76	0.26

VI. Nematode Management

Nematicide Evaluation in a Root-Knot Nematode Infested Field in Central Alabama, 2018

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Biological seed treatments were used for growth and yield evaluations in a root-knot nematode infested soybean field. The biological seed treatments included ALB-206, ALB-305, ALB-BE3, ALB-BE3+SAR, ALB-BE3+EE-FAL, and HM-1805+SAR. All the seeds contained base fungicide and insecticide (F&I) treatments. The experimental treatments contained the base F&I plus additional product. Seeds with base F&I and F&I+AVEO were used as controls in the test. Albaugh, LLC treated all seeds with base F&I and experimentals prior to furnishing the seed for our trial. This trial was conducted at the Plant Breeding Unit near Tallassee, Alabama. This field is naturally infested with root-knot nematode at a large population density. The soil type is Kalmia loamy sand (80% sand, 10% silt and 10% clay). Plots were arranged in a randomized complete block design with five replications. Plots consisted of 2 rows, 7.6 m long with 1 m row spacing, and a 6 m wide alley between the plots. Planting occurred on 4 May. All plots were maintained throughout the season with standard herbicide, insecticide, and fertility production practices as recommended by the Alabama Cooperative Extension System. Plots were irrigated with a lateral line irrigation system as needed. Plant stand was determined in 1.5 m length of row per plot, 12 days after planting (DAP) on 16 May. Samples were collected for nematode analysis and soybean growth measurements by digging 4 random plants per plot after 31 DAP on 13 June. Plant height, shoot and root fresh weight were measured prior to nematode egg extraction. Root-knot nematode eggs were extracted by shaking the roots in a 6% NaOCl solution for 4 minutes and collecting the eggs on a 25 µm sieve. The root-knot nematode population density was determined as the number of root-knot nematode eggs per gram of root. Plots were harvested 158 DAP on 9 October. Data were analyzed with SAS 9.4 using PROC GLIMMIX and LS-means were compared using Tukey-Kramer's method ($P \leq 0.10$). Monthly average maximum temperatures from planting in May through harvest in October were 34, 36, 36, 37, 36, and 34°C with average minimum temperatures of 10, 16, 19, 18, 20, and 5°C, respectively. Rainfall accumulation for each month was 10.3, 9.1, 11.7, 7.6, 4.0, and 10.3 cm with a total of 53.0 cm over the entire growing season.

Average plant stands ranged from 20 to 24 plants per 1.5m of row per plot. ALB-BE3+SAR and HM-1805+SAR treatments had significantly higher stand count than ALB-BE3+EE-FAL treatment. The average plant height of ALB-206, ALB-305, ALB-BE3+SAR, ALB-BE3+EF-FAL, HM-1805+SAR, and AVEO treated seeds were numerically higher than the basic F&I, but statistically the increments were not significant at *(P≤0.1).* ALB-305 and F ALB-BE3+SAR numerically increased the shoot fresh weight up to 20%, but there were no statistical differences. Root fresh weights were significantly increased by ALB-BE3, ALB-BE3+SAR, and HM-1805+SAR compared with all other treatments. Root-knot nematode population density was significantly higher in ALB-BE3+EE-FAL than in ALB-206 and ALB-BE3+SAR. Compared to base F&I treatment, ALB-BE3+SAR, ALB-206, ALB-BE3, HM-1805+SAR, AVEO, and ALB-305 reduced nematode population density by 85%, 76%, 56%, 51%, 40%, and 36% respectively. Soybean yields were not significantly different but numerical variation was observed. The ALB-BE3-SAR, ALB-206, and ALB-BE3 increased the yield by 23%, 18% and 11% respectively.

Treatment	12 DAP	31 DAP				158 DAP
	Plant stand[z]	Plant height[y] (cm)	Shoot fresh weight (g)	Root fresh weight (g)	*Meloidogyne incognita*[x] eggs/g root	Yield (kg/ha)
Fungicide & Insecticide[w]	23 AB[v]	18.6 A	40.4 A	10.1 B	5136 AB	2222 A
ALB - 206	23 AB	19.2 A	39.9 A	11.4 B	1205 B	2624 A
ALB - 305	22 AB	18.9 A	48.4 A	11.2 B	3415 AB	2043 A
ALB - BE3	23 AB	18.6 A	39.2 A	12.7 A	2266 AB	2466 A
ALB - BE3 + SAR	24 A	20.2 A	48.6 A	12.8 A	750 B	2741 A
ALB - BE3 + EE-FAL	20 B	19.2 A	39.7 A	12.0 B	6714 A	2386 A
HM-1805 + SAR	24 A	19.1 A	41.0 A	12.7 A	2496 AB	2184 A
AVEO	22 AB	19.9 A	44.0 A	11.6 B	3080 AB	2102 A

[z] Plant stand was the average number of seedlings in 1.5 m length of row per plot.
[y] Plant height was the average height of four random plants per plot measured in centimeters.
[x] *Meloidogyne incognita* eggs/g of root was the average number of root-knot nematode eggs per gram of root from four root systems.
[w] Fungicide & insecticided treatment include treatment with Vibrance and Cruiser. All other treatments included the fungicide and insecticide plus additional product (s).
[v] Values present are the LS-means separated by Tukey-Kramer method at *P≤0.1.* Values in the same column followed by the same letter do not differ significantly.

Evaluation of BioST Nematicide for Root-Knot Nematode Management on Soybean in Central Alabama, 2018

B. R. Lawaju, K. S. Lawrence, W. Groover, D. Dyer, K. Gattoni, M. N. Rondon, and W. Sancehz

The nematicide BioST, along with ioFRESH, SAR HU, TANNIN, and ISR 2000 were applied as seed treatments for the management of root-knot nematode on soybean in a naturally infested field. Albaugh, LLC treated the seeds with the products prior to providing them for the test. All the seeds contained base fungicide and insecticide (F&I) treatments whereas experimental treatments had base F&I plus the additional product. Seeds with base F&I were used as control. The test was conducted at the Plant Breeding Unit of the E. V. Smith Research Center near Tallassee, AL. The soil is Kalmia loamy sand with 80% sand, 10% silt, and 10% clay. Plots consisted of 2 rows, 7 m long with 0.9 m spacing and were arranged in a randomized complete block design with five replications. Blocks were separated by a 6 m wide alley. All plots were maintained with standard herbicide, insecticide, and fertility production practices throughout the season as recommended by the Alabama Cooperative Extension System. The trial was planted on 4 May. Plant stand was determined at 19 days after planting (DAP) by counting the number of seedlings in 1.5 m of row per plot. Plant height, shoot fresh weight, root fresh weight, and nematode egg counts were measured from 4 random plant samples dug from each plot at 31 DAP. Root-knot nematode eggs were extracted by shaking the roots in 6% NaOCl for 4 minutes and collecting the eggs on a 25 µm sieve. The root-knot nematode population density was determined as the number of root-knot nematode eggs per gram of root. Plots were harvested at 158 DAP. Data were analyzed by ANOVA using PROC GLIMMIX with SAS 9.4 (SAS Institute, Inc., Cary, NC) and LS - means compared with the Tukey-Kramer method at the significant level $p \leq 0.1$. Monthly average maximum temperatures from planting in May through harvest in October were 34, 36, 36, 37, 36, and 34°C with average minimum temperatures of 10, 16, 19, 18, 20, and 5°C, respectively. Rainfall accumulation for each month was 10.3, 9.1, 11.7, 7.6, 4.0, and 10.3 cm with a total of 53.0 cm over the entire growing season.

Plant stand counts were similar among all the treatments and ranged from 21 to 24 plants in 1.5 m length of row per plot. Plant height was also similar across all treatments but shoot and root fresh weights were significantly increased by BioST + ISR2000 compared with most other treatments. Root-knot nematode population density was substantial in all treatments. The root-knot

nematode population density was statistically similar among all the treatments, but numerical difference was observed. The BioST+ SAR HU and BioST + ISR2000 numerically reduced root-knot nematode eggs per gram of root by 44% and 11 % respectively as compared with the F&I treatment. Soybean yields were not significantly different, but all the treatments had numerically higher yield than control except BioST+SAR HU. The BioST + ISR2000 treatment had the highest yield, numerically, followed by BioST, BioST + TANNIN, and BioST + ioFRESH treatments.

Treatment	19 DAP	31 DAP				158 DAP
	Plant stand[z]	Plant height[y] (cm)	Shoot fresh weight (g)	Root fresh weight (g)	*Meloidogyne incognita*[x] eggs/g root	yield (kg/ha)
Fungicide & Insecticide[w]	24 A[v]	19.9 A	45.9 B	10.3 B	2062 A	2377 A
BioST	23 A	20.1 A	51.7 AB	11.3 AB	3149 A	2798 A
BioST + ioFRESH	22 A	20.7 A	49.4 B	11.1 AB	2024 A	2500 A
BioST + SAR HU	21 A	20.4 A	49.2 B	9.3 B	1146 A	2330 A
BioST + TANNIN	21 A	19.9 A	46.4 B	10.5 B	3003 A	2562 A
BioST + ISR 2000	22 A	21.1 A	60.4 A	13.0 A	1846 A	2825 A

[z] Plant stand was the number of seedlings in 1.5 m length of row per plot.
[y] Plant height was the average height of four random plants per plot measured in centimeters.
[x] *Meloidogyne incognita* eggs/g was the average number of root-knot nematode eggs per gram of root from four root systems.
[w] Fungicide & insecticide treatment included treatment with Vibrance and Cruiser. All other treatments included the fungicide and insecticide plus additional product (s).
[v] Values present are LS- means separated by the Tukey-Kramer method at $P \leq 0.1$. Values in the same column followed by the same letter do not differ significantly.

Evaluation of Nematicide Seed Treatments for Management of Reniform Nematode on Soybean in North Alabama, 2018

M.N. Rondon, K. S. Lawrence, W. Groover, D. Dyer, K. Gattoni, B. R. Lawaju, and W. Sanchez

The use of different nematicides were evaluated for reniform nematode management on AG 51X8 soybean at the Tennessee Valley Research and Education Center (TVREC) located near Belle Mina, AL. The soil type was a Decatur silt loam soil with 24% sand, 49% silt, and 28% clay. Seed treatments were applied to the soybean seeds by Bayer CropScience. Seeds were sowed in the field on 08 May 2018. Plots consisted of two rows that were 7.6 meters long with 1-m row spacing, arranged in a RCBD with four replications. All plots were maintained throughout the season with standard herbicide, insecticide, and fertility production practices and a lateral irrigation system was used as needed. Plant stand counts were recorded 16 days after planting (DAP). Plant height, biomass, and nematode population data were collected at 43 DAP. Biomass was calculated as the sum of the root fresh weight and the shoot fresh weight in grams. Nematodes were extracted by soaking the roots in a 6% NaOCl solution on an orbital shaker for 4 minutes, and nematodes were collected on a 25-µm sieve. Plots were harvested on 23 October. Data were analyzed with SAS 9.4 (SAS Institute, Inc., Cary, NC) using PROC GLIMMIX, and means were compared to the control (Gaucho 600) using Dunnett's method with different significant levels. Monthly maximum temperatures from planting in May through harvest in October were 86.2, 90.8, 91.0, 90.8, 89.3, and 77.2 °F with average minimum temperatures of 75.3, 79.8, 80.8, 80.0, 79.4, and 66.1 °F, respectively. Rainfall accumulation for each month was 3.40, 4.47, 2.20, 4.42, 3.09, and 2.44 in., with a total of 20.02 in. over the season.

Plant stand at 16 DAP ranged from 18 to 26 plants per 7.6 meter of row, being statistically lower for all treatments compared to Gaucho 600 (1), which indicated phytotoxicity of the nematicides. Plant height was affected for the nematicides treatments, which was significantly lower for IleVO 600 + Gaucho 600 (3) and IleVO 720 FS + Gaucho 600 (5) compared to Gaucho 600 (1). VOTiVO FS 240 + Gaucho 600 (4) had significantly higher biomass compared to the control, Gaucho 600 (1). IleVO 600 + Gaucho 600 (2) was the only treatment with a significantly lower number of nematode eggs, compared to Gaucho 600 (1). There were no significant differences in soybean yield across the nematicides, but IleVO 600 + Gaucho 600 (2) and IleVO 720 FS + Gaucho 600 (5) had numerically higher yields compared to Gaucho 600 (1).

No.	Seed treatment[y]	Rate	Stand (16 DAP)[z]	Plant height (cm)	Biomass (g)	*Rotylenchulus reniformis* (eggs/g root)	*Rotylenchulus reniformis* (eggs/4 plants)	Yield (kg/ha)
1	Gaucho 600	0.12 mg ai/seed	26	26.68	76.77	317	3030	2841
2	IleVO 600 + Gaucho 600	0.15 mg ai/seed + 0.12 mg ai/seed	21**	24.68	79.08	58*	858*	2890
3	IleVO 600 + Gaucho 600	0.075 mg ai/seed + 0.12 mg ai/seed	21**	23.68*	69.78	175	2156	2511
4	VOTiVO FS 240 + Gaucho 600	3 miu/seed + 0.12 mg ai/seed	18****	26.62	105.63*	197	2740	2744
5	IleVO 720 FS + Gaucho 600	0.15 mg ai/seed + 0.12 mg ai/seed	21**	21.68***	57.77	223	2772	2868
6	IleVO 720 FS + Gaucho 600	0.075 mg ai/seed + 0.12 mg ai/seed	22**	25.87	78.29	128	1968	2717
7	Poncho/VOTiVO	0.13 mg ai/seed	21**	24.37	63.66	258	2926	2650

[y]All seeds were treated with a base seed treatment of Evergol Energy (65 ml/100 kg), Allegiance FL (0.02 mg ai/seed), Precise S Finisher 1010 (65 ml/100 kg), and Pro-Ized Red Colorant (65 ml/100 kg).
[z]Means in the same column followed by * ($P \leq 0.1$), ** ($P \leq 0.05$) ,*** ($P \leq 0.01$) and **** ($P \leq 0.001$) according to Dunnett's P values compared to the control (Gaucho 600 FS) are significantly different.

Soybean Nematicide Combinations for Reniform Nematode Management in Limestone County, 2018

M. N. Rondon, K. S. Lawrence, W. Groover, D. Dyer, K. Gattoni, B. R. Lawaju, and W. Sanchez

Different nematicide combinations were evaluated for reniform nematode management on AG 51X8 soybean at the Tennessee Valley Research and Education Center (TVREC) located near Belle Mina, AL. The soil type was a Decatur silt loam soil with 24% sand, 49% silt, and 28% clay. Seed treatments were applied to the soybean seeds by Bayer CropScience. Seeds were sowed in the field on 08 May 2018. Plots consisted of two rows that were 7.6 meters long with 1-m row spacing, arranged in a RCBD with five replications. All plots were maintained throughout the season with standard herbicide, insecticide, and fertility production practices and a lateral irrigation system was used as needed. Plant stand counts were recorded 16 days after planting (DAP). Plant height, biomass, and nematode population data was collected at 43 DAP. Biomass was calculated as the sum of the root fresh weight and the shoot fresh weight in grams. Nematodes were extracted by soaking the roots in a 6% NaOCl solution on an orbital shaker for 4 minutes, and nematodes were collected on a 25-µm sieve. Plots were harvested on 23 October. Data was analyzed with SAS 9.4 (SAS Institute, Inc., Cary, NC) using PROC GLIMMIX, and means were compared to the control (Gaucho 600) using Dunnett's method with different significant levels. Monthly maximum temperatures from planting in May through harvest in October were 86.2, 90.8, 91.0, 90.8, 89.3, and 77.2 °F with average minimum temperatures of 75.3, 79.8, 80.8, 80.0, 79.4, and 66.1 °F, respectively. Rainfall accumulation for each month was 3.40, 4.47, 2.20, 4.42, 3.09, and 2.44 in., with a total of 20.02 in. over the season.

A low variation of the plant stand at 16 DAP was observed for all treatments, ranging from 20 to 24 soybean plants per 7.6 meter of row. No significant difference was observed in plant height and biomass across the treatments compared with Gaucho 600 (1). BIOst 100 (5) exhibited a numerically higher biomass than other treatments. The use of nematicides on AG 51X8 soybean cultivar exhibited no significant response for the number of nematode eggs in these conditions. Soybean yield was numerically higher when BIOst 100 (5) and Aveo EZ (4) was used to control nematodes, even though these treatments exhibited numerically higher number of nematodes

compared to Gaucho 600. BIOst (5) and Aveo EZ (4) numerically increased soybean yield in 10 and 9% compared to Gaucho 600 (1), respectively.

No.	Seed treatment[y]	Rate	Stand (16 DAP)[z]	Plant height (cm)	Biomass (g)	*Rotylenchulus reniformis* (eggs/g root)	(eggs/4 plants)	Yield (kg/ha)
1	Gaucho 600	0.12 mg ai/seed	24	28.90	72.20	29	351	3794
2	IleVO + Gaucho 600	0.075 mg ai/seed + 0.12 mg ai/seed	23	27.60	80.89	40	327	4059
3	IleVO + Gaucho 600	0.15 mg ai/seed + 0.12 mg ai/seed	20	27.05	81.40	41	498	3942
4	Aveo EZ	0.02 ml/1000 seed	21	29.25	80.62	66	786	4130
5	BIOst 100	195 ml/100 kg	22	29.55	82.27	47	564	4160

[y]All seeds were treated with a base seed treatment of Fluoxastrobin FS480 (7.5 g ai/100 kg), Proline 480 SC (7.5 g ai/100 kg), Allegiance FL (0.02 mg ai/seed), Precise S Finisher 1010 (65 ml/100 kg), Pro-Ized Red Colorant (19.6 ml/100 kg).

[z]Means in the same column followed by * ($P \leq 0.1$), ** ($P \leq 0.05$) ,*** ($P \leq 0.01$) and **** ($P \leq 0.001$) according to Dunnett's P values compared to the control (Gaucho 600 FS) are significantly different.

Soybean Nematicide Combinations for Reniform Nematode Management in North Alabama, 2018

M. N. Rondon, K. S. Lawrence, W. Groover, D. Dyer, K. Gattoni, B. R. Lawaju, and W. Sanchez

Different nematicide combinations were evaluated for reniform nematode management on AG 53X6 soybean at the Tennessee Valley Research and Education Center (TVREC) located near Belle Mina, AL. The soil type was a Decatur silt loam soil with 24% sand, 49% silt, and 28% clay. Seed treatments were applied to the soybean seeds by Bayer CropScience. Seeds were sowed in the field on 08 May 2018. Plots consisted of two rows that were 7.6 meters long with 1-m row spacing, arranged in a RCBD with five replications. All plots were maintained throughout the season with standard herbicide, insecticide, and fertility production practices and a lateral irrigation system was used as needed. Plant stand counts were recorded 16 days after planting (DAP). Plant height, biomass, and nematode population data was collected at 43 DAP. Biomass was calculated as the sum of the root fresh weight and the shoot fresh weight in grams. Nematodes were extracted by soaking the roots in a 6% NaOCl solution on an orbital shaker for 4 minutes, and nematodes were collected on a 25-µm sieve. Plots were harvested on 23 October. Data was analyzed with SAS 9.4 (SAS Institute, Inc., Cary, NC) using PROC GLIMMIX, and means were compared to the control (Gaucho 600) using Dunnett's method with different significant levels. Monthly maximum temperatures from planting in May through harvest in October were 86.2, 90.8, 91.0, 90.8, 89.3, and 77.2 °F with average minimum temperatures of 75.3, 79.8, 80.8, 80.0, 79.4, and 66.1 °F, respectively. Rainfall accumulation for each month was 3.40, 4.47, 2.20, 4.42, 3.09, and 2.44 in., with a total of 20.02 in. over the season.

Plant stand at 16 DAP ranged from 17 to 23 soybean plants per 7.6 meter of row. Two concentration of IleVO combined with Gaucho 600 (2 and 3) and BIOst 100 (5) statistically increased the number of alive plants compared to Gaucho 600 (1). Lower concentration of IleVO + Gaucho 600 (2), Aveo EZ (4), and BIOst 100 (5) increased plant height compared to Gaucho 600 (1). Biomass was higher only for IleVO + Gaucho 600 (3) and Aveo EZ (4) compared with Gaucho 600 (1). Reniform nematode population densities were lower than anticipated ranging from 44 to 51 for the AG 53X6 soybean variety resulting in no significant difference across the treatments. No significant difference was observed for soybean yield, but IleVO + Gaucho 600 at various rates (2 and 3) supported the numerically highest soybean yield. The numerically highest

yield was obtained using IleVO 0.15 mg ai/seed + Gaucho 600 (3) as a nematicide, 6.5% higher than Gaucho 600 (1).

No.	Seed treatment[y]	Rate	Stand (16 DAP)[z]	Plant height (cm)	Biomass (g)	*Rotylenchulus reniformis* (eggs/4 plants)	Yield (kg/ha)
1	Gaucho 600	0.12 mg ai/seed	17	21.30	46.92	44	2837
2	IleVO + Gaucho 600	0.075 mg ai/seed + 0.12 mg ai/seed	23***	24.10*	61.09	51	2995
3	IleVO + Gaucho 600	0.15 mg ai/seed + 0.12 mg ai/seed	22**	23.45	67.21**	41	3021
4	Aveo EZ	0.02 ml/1000 seed	20	24.05*	63.83*	44	2900
5	BIOst 100	195 ml/100 kg	21*	23.85*	53.71	49	2759

[y]All seeds were treated with a base seed treatment of Fluoxastrobin FS480 (7.5 g ai/100 kg), Proline 480 SC (7.5 g ai/100 kg), Allegiance FL (0.02 mg ai/seed), Precise S Finisher 1010 (65 ml/100 kg), Pro-Ized Red Colorant (19.6 ml/100 kg).

[z]Means in the same column followed by * ($P \leq 0.1$), ** ($P \leq 0.05$) ,*** ($P \leq 0.01$) and **** ($P \leq 0.001$) according to Dunnett's P values compared to the control (Gaucho 600 FS) are significantly different.

VII. Extras

Alabama Row Crops Short Course

D. Delaney, B. Ortiz, T. Sandlin, K. Balkcom, A. Gamble, and S. Li

The 2018 Alabama Row Crops Short course took place at the Auburn University Hotel on December 13 and 14, 2018. This was the second year that we included topics related to the major row crops planted in Alabama. The successful event was well-received by the participants. We had approximately 240 participants at this year's Short Course, up from 160 in 2017.

Most of the speakers came from different universities across the south, but also included a "High-yield" farmer panel and representatives from national organizations and the U.S. Congress. The program included two general sessions and 2 crop-related breakout sessions. The program covered a broad variety of topics ranging from tariffs and market outlooks, weed management including auxin herbicides, plant diseases, insect pest issues, fertilization, irrigation water management, cover crops and more. Our hope is that this program can continue to be conducted on an annual basis, and will be of value to farmers and agricultural professionals. Thanks you for your support.

Improving Soil Quality in Alabama

G. Huluka

We provided matching fund for a total of 164 samples during the fiscal year that were sent to the lab. Due to the low number of samples that were submitted to the lab, we covered all the costs associated with the Alabama Soil Quality Index (SQI) test by the Auburn University Soil Testing lab. We have been advertising the availability of financial help to researchers and farmers who are interested for the service. We are optimistic that more soil samples for SQI will be sent to the lab in 2019. Hence, your continued support will be needed as we expect more samples in the near future. An example of Alabama Soil Quality Index report sent out to customers is shown below.

Auburn University Soil Health Index Report

Name: **Your Name** Date of Report: 5/30/2018
Sample #: 20182171 Sample Name: Pasture 2
Crop(s): Pasture

Factor	Values					Max. Score	Your Score	Your Value	Recommendations
Soil CEC/Soil Group	<4.6 (Grp 1)	4.7-9.0 (Grp 2)	9.0-15.0 (Grp 3)	>15 (Grp 4)		5	5	9.51	
Soil pH	Very Acidic	Acidic	Optimal	Alkaline		15	15	6.3	
P Rating	VL/Low	Medium	High	Very High	Extremely High	10	0	VL 4	See soil test P recommendations
K Rating	VL/Low	Medium	High	Very High	Extremely High	10	5	M 158	See soil test K recommendations
Base Saturation	<10%	11-25%	26-50%	50-75%	>75%	10	10	71%	
Soil O.M. %	<1.0	1.0-2.0	2.0-3.0	3.0-4.0	>4.0	20	20	5.1%	
Soil Respriation (Microbial Activity)	Very Low	Low	Moderate	High	Very High	10	4	2.93	PP1, PP2, PP3, PP5, SP7
N Mineralized (lbs/ac/yr)	<10	10-20	20-40	40-80	>80	10	4	20-40	PP1, PP3, PP4
Aggregate Stability	No Aggregates	Weak	Moderate	Strong	Very Strong Aggregates	10	2		PP1, PP2, PP3, SP7, SP2
TOTAL SOIL QUALITY INDEX:						**100**	**65**		See BMPs Above

Comments: Soil could use improvement. Consider implementing one or more of the above practices.

* Increasing soil organic matter will also help increase CO_2 respiration (an indicator of microbial activity), plant-available nitrogen (through microbial breakdown of organic matter), and aggregate stability.

***"This Soil Quality Test has been run at a 50% discount because of checkoff support from* Alabama Soybean Committee."**

Detection of Cassicolin-Encoding Genes in *Corynespora cassiicola* Isolates from Soybean and Cotton

K. S. Lawrence, J. Koebernick, and M.N. Rondon

Justification: *Corynespora cassiicola* is a fungal pathogen with increasing importance across soybean and cotton producing countries and it is responsible for target spot disease in these crops. A small protein named cassiicolin produced by *C. cassiicola* isolates has been reported as an essential effector for the pathogenicity. It is difficult to develop resistant cultivars by breeding programs without understand the pathogenicity of the *C. cassiicola* isolates in our region. As target spot becomes more relevant as a plant disease due to its increasing occurrence on several high economic value crops, it is important to gain more understanding about its pathogen, *Corynespora cassiicola* and its toxin, cassiicolin. Therefore, isolates from soybean and cotton could be separated by pathogenicity and could be used in breeding for screening lineages. Thus, the mains aim of this project is to detect the cassiicolin-encoding genes (*Cas1* to *Cas6*) from several *C. cassiicola* isolates from cotton and soybean in Alabama.

Objective: The mains objective of this project is to detect the cassiicolin-encoding genes (*Cas1* to *Cas6*) from multiple *C. cassiicola* isolates from soybean and cotton in Alabama. Soybean and cotton target spot symptomatic leaves will be collected from different locations of Alabama. *Corynespora cassiicola* isolates collected from the samples will be identified by morphological and molecular characters. In sequence, all isolates obtained will be submitted to DNA extraction and PCR identification with specific primers covering *Cas* sequences for gene detection.

Results: Our preliminary results indicate an important genetic diversity of *C. cassiicola* isolates sampled in Alabama U.S. based on the cassiicolin-encoding genes, being higher for isolates sampled from soybean. Twelve of the *C. cassiicola* isolates from soybean indicate they have co-existence of different Cas genes (Cas2+6).

Outcome: Our findings will help assess soybean susceptibility or resistance to *C. cassiicola* based on the toxin encoding genes in variety evaluations and a soybean breeding program.

AMOUNT REQUESTED: Total costs that will be $10,000

1. Salaries
2. Wages
3. Graduate student (1/3) time - $7,500 (Marina Rondon)

1. Benefits- $ 135

4. Operating
    a. Travel -$2,365
        i. Trips to the TVREC, GCREC, PBU, EVS, Brewton, Prattville and growers fields…. mileage and meals each and we expect to monitor the fields monthly during the season.

Detection of Cassicolin-Encoding Genes in *Corynespora cassiicola* Isolates from Soybean and Cotton

K. S. Lawrence, J. Koebernick, and M.N. Rondon

Results: Cotton and soybean isolates fell into different profile clusters for cassiicolin-encoding genes in 191 *C. cassiicola* isolates (Table 1). Two cassiicolin-encoding clusters of genes from cotton isolates were found (*Cas2* and *Cas2+6*), while three cassiicolin-encoding clusters of genes from soybean isolates were found (*Cas2*, *Cas6* and *Cas2+6*). Seven cotton isolates and 10 soybean isolates did not exhibited the presence of any cassiicolin-encoding genes (Cas0). Two cassiicolin-encoding genes (*Cas2+6*) were found together for 81 isolates sampled from soybean and only one from cotton.

Of 66 cotton isolates and 125 soybean isolates assayed, 58 (87.9%) cotton isolates, and only 28 (22.4%) soybean isolates were positive for the *Cas2* gene by PCR detection. While 81 (64.8%) isolates from soybean were positive for the combination *Cas2+6* genes, but only one isolate from cotton (1.5%). Therefore, *Cas2* was the dominant gene for cotton isolates, and the combination *Cas2+6* was the dominant gene for soybean isolates. *Cas6* gene alone were found in only six soybean isolates (4.8%), while Déon et al. (2014) did not find any isolate with *Cas6* alone even assessing 70 isolates from most of the *C. cassiicola* host plants.

The PCR products obtained from genomic DNA of the *C. cassiicola* isolates from cotton (HSV 01, FHP 22, BRW 03, EVS 01, HSV 12 and FHP 01) and soybean (LIM 02, PBU 06, LIM 14, PBU 07, LIM 13 and PBU 04) used for illustration here represent 6 isolates from each host (Figure 1). The primers covering the *Cas* sequences were able to amplify fragments around 750 bases of pairs on agarose gels. No amplification product was obtained from the control, which water was used instead of DNA.

The diversity of the cited genes with different forms and combinations of the cassiicolin toxin found for cotton and soybean isolates in eight Alabama counties (Figure 2) could explain some symptoms differentiation. Further work assessing the pathogenicity, the cultivar preferences, severity from isolates genetically distinct based on the toxin when inoculated on a range of cultivars is required.

Our results indicate an important genetic diversity of *C. cassiicola* isolates collected in Alabama, U.S. based on the cassiicolin-encoding genes, being higher for isolates from soybean. Additional sampling of isolates from cotton may reveal higher diversity. Our findings may help to develop

ways to assess susceptible and resistant cultivars to *C. cassiicola* in a breeding program by testing genetically distinct isolates.

Table 1. *Corynespora cassiicola* isolates from cotton and soybean with the corresponded target gene.

Target gene	*Corynespora cassiicola* isolates		Total
	Cotton	Soybean	
Cas0	7	10	**17**
Cas2	58	28	**86**
Cas6	0	6	**6**
Cas2+6	1	81	**82**
Total	**66**	**125**	**191**

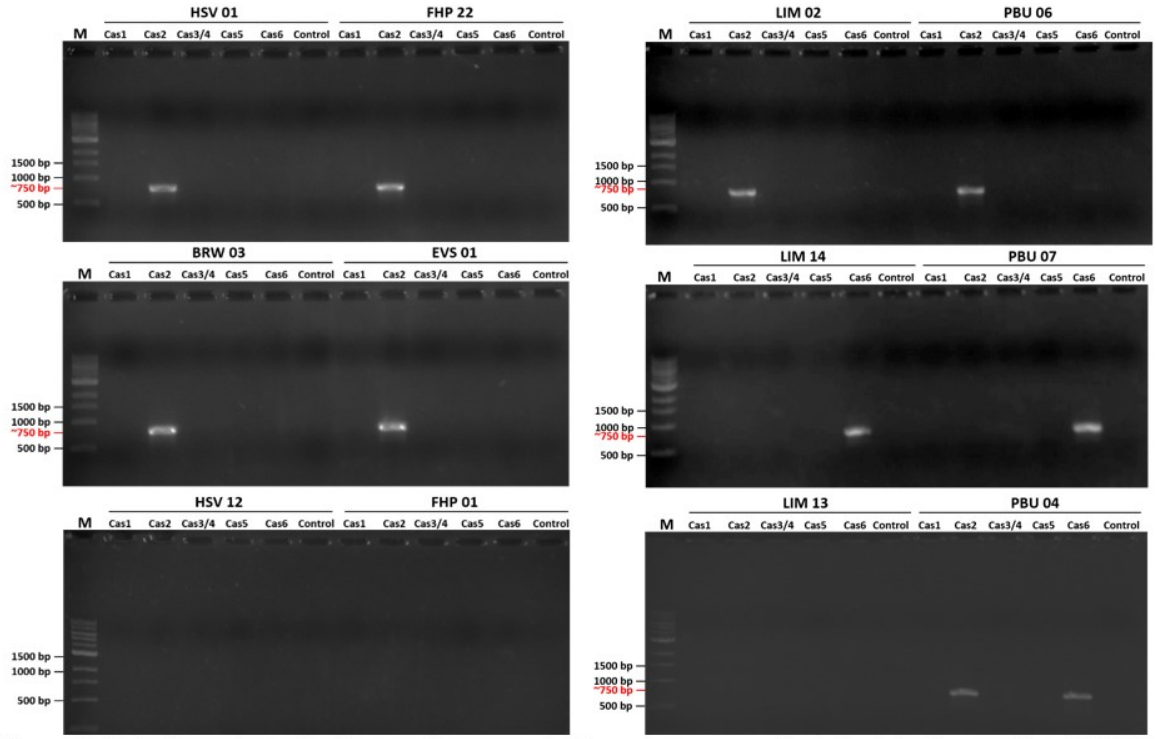


Figure 1. Detection of cassiicolin-encoding genes in *Corynespora cassiicola* isolates from cotton (HSV01, FHP22, BRW03, EVS01, HSV12, and FHP01), at the left; and isolates from soybean (LIM02, PBU06, LIM14, PBU07, LIM13, and PBU04), at the right. M, molecular weight marker; Control, water (without DNA extract).

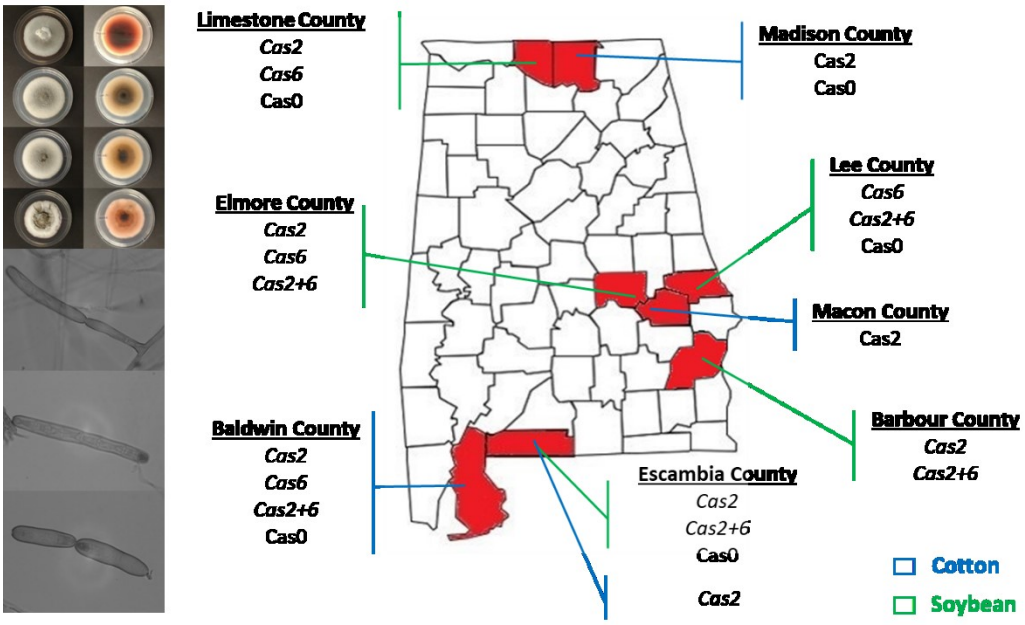


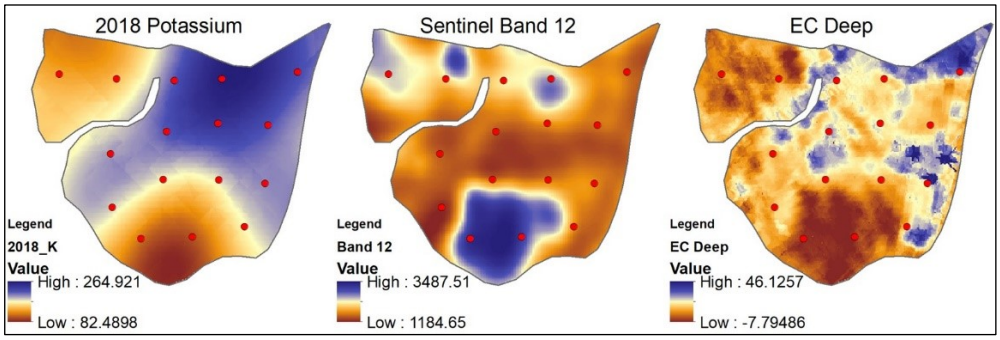
Figure 2. Distribution map of the cassicolin-encoding genes found in isolates sampled from cotton (blue colored) and soybean (green colored) in Alabama, U.S.

Development of Soil Ssampling Zones Using Remote Sensing

B. Ortiz and J. Oldag

While the industry standard for soil sampling intensity remains a 2.5 acre grid, many farmers and consultants are beginning to find that this uniform approach to sampling does not always provide the information needed for precision agriculture. The main goal of this project is to identify whether or not the 2.5 ac sampling grid is still valid or a different approach should be implemented, especially if sampling-design choices can be guided by the use of ancillary data. The first objective of this project is to confirm that greater sampling intensity is often needed to accurately represent in-field soil variability for precision agriculture purposes. This project was initiated in 2017 and continue 2018 with data analysis and collection at two farms, L. C. farms in Samson, AL and Autauga Farming Co. in Autaugaville, AL.

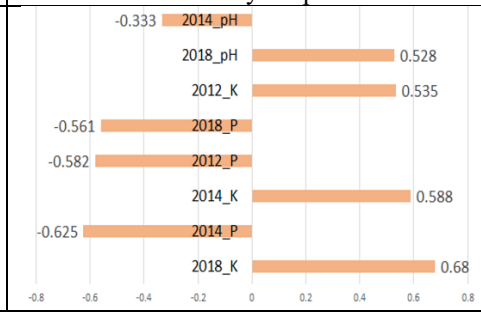
The 2018 activities were focused on the evaluation of multiple datasets for their use on guiding soil sampling for soil macronutrients and soil pH. The maps below shows the spatial correlation (agreement) between the changes in 2018 Potassium and data from a Sentinel satellite image and also soil electrical conductivity (Soil EC_a) . Results also indicated that high soil EC values could be associated with high soil K values and vice versa occurs for soil P. The dots on the maps correspond to the 2.5 ac standard sampling used by a consultant hired by the farmer. Results indicated that either soil EC_a or the image can be used to determine where to collect samples and in the case of this field, zone sampling is a better approach.



Correlation of soil macronutrients and pH with data from satellite images (2018)

Nutrients	Ancillary Data	Correlation
2018_K	Sentinel Band 12	-0.548
2018_K	Sentinel Band 11	-0.519
2018_P	Sentinel Band 12	0.387
2018_pH	Sentinel Band 02	0.369
2018_P	Sentinel Band 11	0.346

Correlation of soil macronutrients and pH with soil electrical conductivity deep



Correlated values suggest that variables vary in the same range. Variables that exhibit the same spatial range with sampled soil variables can be used for the prediction of soil macronutrient

sampling zones. By creating zones delineated by ancillary data (images or soil ECa), areas of high and low variability can be identified. These zones of high variability indicate a greater need for intense soil sampling, while low variability requires less sampling. These zones can be used to guide thorough and efficient soil sampling processes. Analyses will continue in 2019.

Support for Precision Agriculture Extension Programs

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Adoption of PA technologies and practices will increase as a result of trainings and on-farm demonstrations. As part of this efforts, we have been trying to establish the Alabama Precision Agriculture Learning network to support training and adoption of technologies and practices. Most of the Precision Ag. extension activities conducted in 2018 were focused on Irrigation. The establishment of two NRCS funded grants to conduct on-farm demonstration projects of soil-sensor based irrigation scheduling and variable rates irrigation occupied almost of the time of my Precision Ag team. Four irrigation on-farm demonstration sites were established in 2018, three in North Alabama and one in Samson Alabama. Four farmer focus groups were established at each demonstration site to train farmers on the use of those technologies and irrigation water management. An Irrigation E-newsletter was also initiated in 2018. Tables 1 and 2 list the meetings, field days, and on-farm demonstrations organized at each location.

Table 1. Precision Ag. trainings conducted in 2018.

Topics	**Workshop location**	**Tentative date**	**No. of Participants**
Assessment of Irrigation needs and project initiation meeting	Belle mina, AL	February 4th	50
Assessment of Irrigation needs and project initiation meeting	Tanner, AL	February 5th	20
Assessment of Irrigation needs and project initiation meeting	Town Creek, AL	February 5th	30
Irrigation field day	Samson, AL	August 28th	60
Preliminary results of demonstrations in 2018 – Town Creek	Town Creek, AL	August 31st	30
Preliminary results of demonstrations in 2018 – Tanner	Tanner, AL	August 30th	15

Table 2. On-farm demonstrations conducted in 2018.

Topics	**Location**
Variable Rate seeding	E.V. Smith research center
Sensor-based irrigation scheduling	Tanner, AL
Sensor-based irrigation scheduling	Athens, AL
Sensor-based irrigation scheduling and Variable Rate Irrigation	Town Creek, AL
Sensor-based irrigation scheduling and Variable Rate Irrigation	Samson, AL

In addition to these activities, we were also invited to give Precision Ag. related presentations at national and international meetings.