North Dakota Soybean Council Management of Soybean Aphids and Interaction with Soybean Cyst Nematode Technical Report – June 30, 2018

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The overall goal of this work is to provide soybean producers with the integrated pest management (IPM) strategies to control these two economic pests: soybean aphids and soybean cyst nematode (SCN).

Research Objectives included:

- To evaluate different insecticide strategies and *Rag* aphid resistant soybean varieties for management of the soybean aphid in ND. Research will demonstrate to producers which insecticide strategy (seed treatment versus foliar applied insecticides), and *Rag* aphid resistant soybean are the most effective for management of soybean aphids. Two application timings were also tested for the foliar-applied insecticides: an early R1 (beginning bloom) and the economic threshold (ET = an average of 250 aphids per plant).
- To study the interaction between soybean aphids and soybean cyst nematode (SCN), specifically how soybean aphids might affect SCN reproduction, and to measure corresponding yield differences for SCN resistant and susceptible varieties.
- 3) To survey populations of soybean aphids for development of insecticide resistance in eastern North Dakota.

<u>Objective 1</u>: Insecticide seed treatment, foliar-applied insecticide and *Rag* aphid resistant soybean varieties

Materials and Methods

Six treatments were tested for soybean aphid control in trials at three locations in eastern North Dakota with a history of high soybean aphid pressure (Casselton, Harwood and Emerado). Treatments included 1) bare seed, 2) Apron Maxx fungicide seed treatment, 3) Cruiser Maxx insecticide seed treatment, 4) Apron Maxx plus Warrior II applied at the R1 growth stage, 5) Apron Maxx plus Warrior II applied at economic threshold (ET), and 6) Cruiser Maxx plus Warrior II applied at ET. Four treatments using an experimental *Rag2* line were discarded because the line did not exhibit resistance to soybean aphids.

Trials were planted in a randomized complete block design with four replications (Table 1). Plots were 10 feet wide (four 30-inch rows) by 20 feet long and seeded at a rate of 150,000 plants per acre. Foliar insecticides were applied at 20 gpa using a tractor-mounted CO_2 boom with TeeJet 11002 nozzles.

Soybean aphids were sampled by counting the number of aphids on 10 randomly selected plants per plot. The mean number of aphids per plot was calculated and used to calculate cumulative aphid days (CAD). Aphid sampling began at first detection and continued weekly through the R6 soybean growth stage. Plots were harvested at maturity using a Hege plot combine (Table 1). Plot yield was calculated and adjusted to 13.5% grain moisture. All data were analyzed using the GLM procedure in SAS statistical software.

Table 1. Dates of planting and harvest for Objective 1.				
Location Planting Harvest				
Emerado	26-May	3-Oct		
Casselton	19-May	12-Oct		
Harwood	20-May	13-Oct		

Results and Discussion

At Emerado (Table 2) where soybean aphid pressure was high, the bare seed and Apron Maxx treatments (non-insecticide treatments) had significantly more CAD than all treatments that received an insecticide. Apron Maxx plus Warrior II applied at R1 had significantly more CAD than all other insecticide treatments . Cruiser Maxx alone, Cruiser Maxx plus Warrior II applied at ET, and Apron Maxx plus Warrior II applied at ET were not significantly different for CAD. For yield at Emerado, the bare seed, Apron Maxx alone and Apron Maxx plus Warrior II at R1 had significantly lower yield than all other treatments. There were no significant Yield differences among Cruiser Maxx alone, Cruiser Maxx plus Warrior II at ET, and Apron Maxx plus Warrior II at ET.

At Casselton (Table 3) where the ET was reached but not exceeded, there were significant differences among treatments for CAD, but there were no significant differences among treatments for yield. CAD were not great enough to cause yield loss. At Harwood (Table 2) where the ET was not reached, there were no significant differences among treatments for CAD and yield.

Results from Objective 1 support the following recommendations to ND soybean producers: The use of an insecticide seed treatment, either with or without a foliar insecticide application at ET, did not provide added yield benefit under low or high aphid pressure compared to non-insecticide treated seed with a foliar insecticide application at the ET. This suggests that the most economically sound strategy for producer is to not use an insecticide seed treatment for soybean aphid control.

A foliar pyrethroid insecticide application at R1 did not prevent aphids from exceeding the ET and causing significant yield loss. In this situation, a producer would have to make another costly foliar insecticide application to avoid yield loss. This is especially important in light of pyrethroid resistance. Even partially resistant aphids could recolonize due to lower potency of the insecticide's residual activity over a short period of time. This strategy of applying a foliar insecticide at R1 could also contribute to more rapid pyrethroid resistance in soybean aphid populations. Early foliar insecticide applications for soybean aphid management are economically unsound, and can contribute to the rapid development of insecticide resistance in soybean aphid populations. Late season soybean aphid population growth should be monitored carefully to avoid unnecessary foliar insecticide applications. Our results continue to support the use of the ET for optimal soybean aphid management and returns.

Table 2. Treatment means for cumulative aphid days (CAD) and yield at Emerado (high aphid pressure), 2017.

Treatment	CAD ¹	Yield ¹ (bu/acre
Bare Seed	14,937a	30.2b
Apron Maxx	12,807b	32.6b
Apron Maxx + Warrior II R1	6,9391c	33.7b
Apron Maxx + Warrior II ET	3,043d	39.6a
Cruiser Maxx	2,215d	39.3a
Cruiser Maxx + Warrior II ET	2,244d	37.8a

¹Means within a column that share the same letter are not significantly different.

Table 3. Treatment means for cumulative	aphid days (CAD) and	yield at Casselton	(ET reached
but not exceeded) and Harwood (ET not re	eached) in 2017.		

Casselton		Harwood			
		Yield ¹			Yield ¹
Treatment	CAD ¹	(bu/acre	Treatment	CAD ¹	(bu/acre
Bare Seed	3,162a	51.3a	Bare Seed	397a	35.4a
Apron Maxx	3,444a	50.3a	Apron Maxx	523a	35.3a
Apron Maxx + Warrior II	2,307b		Apron Maxx + Warrior II	394a	
R1		51.5a	R1		36.1a
Apron Maxx + Warrior II	2,194bc		Apron Maxx + Warrior II	387a	
ET		52.5a	ET		35.7a
Cruiser Maxx	1,603c	52.4a	Cruiser Maxx	397a	35.9a
Cruiser Maxx + Warrior II	2,097bc		Cruiser Maxx + Warrior II	364a	
ET		51.1a	ET		34.8a

¹Means within a column that share the same letter are not significantly different.

Objective 2: Soybean Aphid & SCN

Materials and Methods

Trials were conducted at two locations in southeastern ND with history of high SCN pressure. The soybean varieties used were Peterson Farms 12R05 (SCN susceptible) and 15R05N (SCN resistant). These varieties were chosen because of their similar maturity and performance in past yield trials. Trials were planted in a randomized split-plot design with four replications (Table 4). Soybean variety was treated as the main plot. Each main plot was divided into three soybean aphid management subplots: 1) soybean aphid free, 2) treatment at ET and 3) no soybean aphid treatment. Main plots were 30 feet wide (12 30-inch rows) by 14 feet long, and each subplot was 10 feet wide (four 30-inch rows). All seed was treated with ApronMaxx fungicide.

Soybean Cyst Nematode was sampled by taking 10 soil cores (five from each of the center two rows) from each subplot. Samples were made in June immediately after seeding, and again in September just prior to harvest. SCN samples were processed by the NDSU Plant Diagnostic Laboratory. Soybean aphids were sampled, and foliar insecticide applications were applied as described in Objective 1. Plots were harvested at maturity using a Hege plot combine (Table 4). Plot yield was calculated and adjusted to 13.5% grain moisture. Data were analyzed using the GLM procedure in SAS statistical software.

Table 4. Dates of planting and harvest for Objective 2.			
Location Planting Harvest			
Colfax	13-May	13-Oct	
Prosper	6-May	16-Oct	

Results and Discussion

Soybean aphids barely reached and did not exceed the ET at Colfax. Soybean aphids did not reach ET at Prosper, most likely due to very dry conditions at the Prosper site. The Prosper site also experienced dicamba drift injury, and this may have contributed to low yields in addition to the drought conditions.

At Colfax (Table 5), there were no significant differences among treatments for June SCN eggs, which indicates that all treatments began with approximately the same SCN level and that SCN was evenly distributed across the trial. For September SCN eggs, 12R05 had significantly more SCN than 15R05N. Among subplots within 12R05, the aphid free subplots had significantly more SCN than the untreated sublpots. There were no significant differences among subplots within 15R05N for September SCN eggs.

For CAD, there were no significant differences among the ET and untreated subplots within and between soybean varieties. This indicates that soybean aphids were evenly distributed across the trial. Comparing CAD with September SCN egg counts within 12R05, we found that the aphid free subplot had the highest SCN count. The ET subplot, while having significantly more CAD than the aphid free subplot, did not differ significantly for SCN eggs. The untreated subplot was not significantly different from the ET subplot for CAD or SCN eggs. Because of the weak interaction between soybean aphid treatment and September SCN egg counts, it is difficult to draw any meaningful conclusions, although our data indicate that reducing soybean aphids results in an increase in SCN when using a SCN susceptible soybean variety. However, this interaction was not reflected in the yield means for the 12R05 subplots, and this interaction was not apparent in 15R05N (the SCN resistant variety). For yield, 15R05N had significantly greater yield compared to 12R05. This is not surprising due to the high SCN pressure.

At Prosper (Table 6), there were no significant differences among treatments for June SCN eggs, which indicates that all treatments began with approximately the same SCN level and that SCN was evenly distributed across the trial. Initial SCN pressure was much higher at Prosper than at Colfax. For September SCN eggs, 12R05 had significantly more SCN than 15R05N. CAD were not significantly different among all treatments, and were too low to make meaningful comparisons between CAD and SCN. Observed yield differences are due to SCN pressure.

Our data suggest that keeping soybeans free of soybean aphids can lead to an increase in SCN. This practice should be avoided and soybean producers should continue to follow the ET guidelines for soybean aphid management whether SCN is present or not. Soybean producers should continue to monitor their fields for SCN, and continue to use SCN integrated pest management tactics, such as SCN resistant varieties.

Table 5. Treatment means for	SCN egg counts per 1	100 cc soil for June a	nd September, and
yield at Colfax, 2017.			

			Cumulative	
	June	September	Aphid	Yield ¹
Treatment ²	SCN Eggs ¹	SCN Eggs ¹	Days ¹	(bu/acre
12R05 Aphid Free	713a	6,876a	390b	33.2c
12R05 ET	590a	5,817ab	1,611a	30.6cd
12R05 Untreated	523a	3,396b	1,666a	26.9d
15R05N Aphid Free	360a	132c	234b	58.1a
15R05N ET	603a	219c	1,137a	53.7ab
15R05N Untreated	913a	228c	1,473a	49.9b

¹Means within a column that share the same letter are not significantly different. ²12R05 is the SCN susceptible variety and 15R05N is the SCN resistant variety.

Table 6. Treatment means for	SCN egg counts per	100 cc soil for June	and September, and
yield at Prosper, 2017.			

	_		Cumulative	
	June	September	Aphid	Yield
Treatment ²	SCN Eggs ¹	SCN Eggs ¹	Days ¹	(bu/acre
12R05 Aphid Free	3 <i>,</i> 060a	4,563a	141a	18.3b
12R05 ET	3 <i>,</i> 000a	4,734ab	267a	17.8b
12R05 Untreated	3 <i>,</i> 355a	3,285b	257a	18.6b
15R05N Aphid Free	4 <i>,</i> 067a	1,650c	136a	20.5a
15R05N ET	3,330a	1,173c	189a	20.4a
15R05N Untreated	3,455a	1,545c	246a	20.4a

¹Means within a column that share the same letter are not significantly different. ²12R05 is the SCN susceptible variety and 15R05N is the SCN resistant variety.

Objective 3: Screening populations of soybean aphids for insecticide resistance

Insect resistance to pesticide is a worldwide problem; the United Nations Environmental Program has listed pesticide resistance as the third most serious threat to global agriculture behind soil erosion and water pollution (Pimentel 2005). In the United States, annual losses due to pesticide resistance are estimated at \$1.4 billion (Oerke 2005). Resistance influences pesticide application costs and crop yields, affecting the level and stability of farm income (Knight and Norton 1989). Several pyrethroid and organophosphate insecticides are commonly used as foliar applications to manage soybean aphids and insecticides continues to be the primary control strategy throughout the Upper Midwest (Hodgson et al. 2012). Soybean aphids could develop insecticide resistance to these chemistries. Based on the 2012 pesticide survey in ND (Zollinger et al. 2014), the top four insecticides used for insect control in soybeans included:

- Pyrethroids: lambda-cyhalothrin (Warrior and generics); bifenthrin (Brigade, Tundra, etc.); esfenvalerate (Asana), and
- Organophosphates: chlorpyrifos (Lorsban and generics).

In Minnesota and Iowa, Hanson et al. (2017) found that soybean aphid populations sampled from 2015 to 2016 had resistance ratios up to 40-fold for pyrethroids (i.e., bifenthrin and lambda-cyhalothrin). As result of these findings, the goal of this research was to survey for populations of soybean aphids in eastern ND that may be developing insecticide resistance using a standardized laboratory bioassay.

Materials and Methods

Populations of soybean aphids were randomly collected from 6 soybean fields that were identified with pyrethroid failure in eastern ND. Collected aphids were taken back to the laboratory for insecticide resistance testing.

A standardized glass-vial bioassay (Figure 1) was used to test two pyrethroid insecticides, bifenthrin and lambdacyhalothrin, for aphid resistance. Each assay consists of three replications of three insecticide concentrations in 20-ml glass vials that have been coated internally with a



Figure 1. Glass-vial bioassay setup for testing pyrethroid resistance to soybean aphids.

solution of technical grade insecticide diluted in acetone at concentrations expected to provide 99% mortality(LC99), twice the concentration expected to provide 99% mortality (2 x LC99), and an acetone control (Table 6).

Table 6. Dosage of insecticide active ingredient			
_		Lambda-	
Dose	Bifenthrin	cyhalothrin	
	μg Al/Vial	μg Al/Vial	
2x99	0.042472866	0.50255892	
1099	0 021236433	0 25630505	

Only wingless adult aphids were used in the bioassay. These aphids were gently transferred with a paint brush into petri dishes containing a piece of filter paper dampened with deionized water. This initial transfer serves to narrow the transfer window into vials allowing all aphids to enter all vials at approximately the same time, and provides a visual assessment of aphid health. Then, aphids were transferred from petri dishes to vials. While wearing nitrile gloves, 10 healthy wingless adult aphids were placed into each treatment vial. Aphids were transferred into treated vials with a specially designated paint brush for each chemical to avoid contamination. Aphids were deposited at the bottom of the vial. Aphids were transferred into all control vials first (Reps 1, 2, and 3), then LC99 vials (Reps 1, 2, and 3), and lastly 2xLC99 vials (Reps 1, 2, and 3) to avoid contamination from a higher concentration. Once infested, vials were kept upright and tightly capped in a tray at 25 C, 70% relative humidity and a 16:8 (L:D) h photoperiod for the duration of the assay.

Mortality of 10 wingless (apterous) adult soybean aphids per vial was assessed after 4 hours and 24 hours of exposure to the inner surfaces of the vials. Mortality of the aphids in vials used the following rating:

- Live: Healthy aphids have a tendency to climb up the walls of the vial and are typically found circling the walls of the vial.
- **Moribund:** Aphids showing a lack of coordinated movement, unable to right themselves, or unable to climb the walls of the vial after 10 seconds should be considered moribund.
- **Dead:** Aphids showing no movement are to be considered dead.

Any aphids that dies for other reasons were recorded, such as parasitized by wasp, infected with mold, found to be have wings, or physical injury during the transfer process. The number of aphid nymphs produced at both 4 and 24 hrs. also were recorded.

Data were analyzed using biases-reduce general linear model with binomial response for Henderson-Tilton-adjusted mortality.

Results and Discussion

In Figure 2, counties highlighted in red are those from which growers reported pyrethroid performance issues for soybean aphid management in 2017. A total of nine counties were reported in eastern ND. Soybean aphids were collected from soybean fields with reported pyrethroid failure, and these aphids were evaluated for bifenthrin and lambda-cyhalothrin resistance using the glass-vial bioassay. Aphid populations from Casselton, Hope, Emerado, Loderma and Osnabrock had significantly less mortality than the laboratory population for bifenthrin (Figure 3a); and Grafton (2 fields), Loderma and Osnabrock for lambda-cyhalothrin (Figure 3b).



Overall, laboratory bioassays confirmed that 83% and 56% of the soybean aphid populations were resistance to bifenthrin and lambda-cyhalothrin, respectively. Pyrethroid resistant soybean aphids will complicate insecticide management decisions for soybean aphids, and create a new challenge for soybean production



Figure 3. 3a. Results of bioassay for bifenthrin and 3b for lambda-cyhalothrin. North Dakota counties are highlighted in yellow, and red bracket indicates sites that have significantly lower mortality compared to the laboratory reared soybean aphids (control).



Outputs:

Information on the results of this research project was disseminated through a new extension publication, *Crop & Pest Report*, and a series of extension or commodity meetings including the 2017 NDSU / UM Commercial Pesticide Applicator Training, *2018 Getting It Right in Soybean Production* meetings and others.

Extension Publication: As result of the research on the development of insecticide resistance in soybean aphids in the upper Midwest, a new multistate extension publication was written and published in February 2017. The publication was a collaborative effort among Extension Entomologists from the University of Minnesota, Iowa State University, NDSU and South Dakota State University.

In 2017, failures of certain pyrethroid insecticides for management of some soybean aphid populations were observed in commercial fields, and were identified as resistant to bifenthrin and lambda-cyhalothrin through small-plot research and laboratory bioassays. This new publication summarizes key pest management recommendations to North Dakota soybean farmers to mitigate the development of insecticide-resistant in populations of soybean aphids:



- Use the economic threshold (E.T.) of 250 aphids per plant with >80% of plants infested to decide when to treat fields.
- Apply insecticide using the full labeled rate with proper spray technology (good coverage) and under favorable environmental conditions (winds <10 mph).
- If more than one application is necessary for soybean aphids or other insect pests, rotate to a different mode of action (MOA).

In E1879, two foliar-applied insecticide tables are included that list the different MOAs for insecticides with a single active ingredient and for premix insecticides with two active ingredients.

The "Management of insecticide-resistant soybean aphids E1878" is available online at <u>https://www.ag.ndsu.edu/publications/crops/management-of-insecticide-resistant-soybean-aphids/e1878.pdf</u>

This work was supported by the North Central Soybean Research Program and each state's soybean society including the Minnesota Soybean Research & Promotion Council, Iowa Soybean Association, North Dakota Soybean Council and South Dakota Soybean Research & Promotion Council.

<u>Peer-reviewed paper</u>: The Journal of Integrated Pest Management (Recommendations section) recently accepted our multi-state manuscript titled **Management of Insecticide-Resistant Soybean Aphids in the Upper Midwest of the United States**. The abstract is listed below:

Management of Insecticide-Resistant Soybean Aphids in the Upper Midwest of the United States Robert L. Koch¹, Erin W. Hodgson², Janet J. Knodel³, Adam J. Varenhorst⁴, and Bruce D. Potter⁵

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ABSTRACT

Since the first observation of soybean aphid in North America in 2000, it has become the most economically damaging insect of soybean in the Upper Midwest of the United States. For the last 17 years, soybean aphid management has relied almost entirely on the use of foliar-applied broad-spectrum insecticides. However, in 2015 in Minnesota, failures of foliar-applied pyrethroid insecticides were reported and pyrethroid resistance was confirmed with laboratory bioassays using lambda-cyhalothrin and bifenthrin. In 2016 and 2017, further reports of failures of pyrethroid insecticides and/or laboratory confirmation of resistance occurred in Iowa, North Dakota, South Dakota, and Manitoba. In response to the challenge posed by insecticide-resistant soybean aphids, we recommend several management strategies for minimizing further development of resistance and subsequent pest-induced crop losses: 1) scout and use the economic threshold to determine when to apply insecticides, 2) apply the insecticides properly, 3) assess efficacy 3-5 days after application, and 4) alternate to a different insecticide group if another application is required. In the long term, soybean aphid management must move beyond insecticide-based management to true integrated pest management by incorporating multiple tactics.

Meetings:

- 2018 Eastern Crop & Pest Management School, February 27-28, 2018, Fargo, ND. Total audience = 96 people.
- The International Crops Expo, February 21-22, 2018, Grand Forks, ND. Total audience = 175 people.
- The Corn-Soybean Expo, February 13, 2018, Fargo, ND. Total audience = >450 people.
- The Best of the Best in Wheat and Soybean Research and Marketing, February 1, 2018, Moorhead, MN. Total audience = 230 people.
- The Best of the Best in Wheat and Soybean Research and Marketing, January 31, 2018, Grand Forks, ND. Total audience = 280 people.
- Getting it Right Soybean Production Meetings 2018. January 26, 2018. Langdon, ND. Total audience = 45 people.
- Getting it Right Soybean Production Meetings 2018. January 25, 2018. Rugby, ND. Total audience = 44 people.

- Getting it Right Soybean Production Meetings 2018. January 24, 2018. Kenmare, ND. Total audience = 70 people.
- Getting it Right Soybean Production Meetings 2018. January 23, 2018. Fessenden ND. Total audience = 41 people.
- NDSU / UM Commercial Pesticide Applicator Training at Fargo Nov. 29, 2017. Total audience = >320 people.
- Field Scouting for Insect Pests of Field Crops, Field tour at NDSU campus, ND State College of Science and Bismarck State College, Fargo, ND – July 21, 2017. Total audience = 75 people.
- Update on Soybean Aphid and Other Crop Pests, NDSU Agronomy Seed Farm Field Tour, Casselton, ND July 17, 2017. Total audience = 60.

An <u>Extension Impact Statement</u> was prepared titled **Managing a New Threat to Soybean Production in ND: Insecticide Resistant Soybean Aphids** for the state legislators. It is available at: <u>https://www.ag.ndsu.edu/impactstatements/impact-statements/2018-impact-statements/2018-impact-statements/18state-knodel-crop.pdf/view</u>

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