Breeding of Glyphosate-Tolerant Soybean Cultivars (2024)
Principal Investigator: Carrie Miranda, PhD

a. Title: Breeding of Glyphosate-Tolerant Soybean Cultivars (2023)
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b. Research Overview: The grant enables NDSU to continue the process of developing glyphosate-tolerant soybean cultivars. This project will significantly reduce seed costs to soybean growers and provide a mechanism for weed management. Glyphosate-tolerant experimental lines in the NDSU breeding program are being tested in a range of maturity that varies from MG 00.6-1.3 and are properly adapted throughout the entirety of North Dakota. New experimental lines are developed each year.

   Objectives: The objective is to continue the process of developing glyphosate-tolerant experimental lines with a goal of releasing cultivars of varying maturities that are developed by NDSU. In addition, other herbicide resistance traits are being explored to incorporate into this project.

c. Materials and Methods:

   The glyphosate tolerant project will end 6/30/2024. It is being modernized as a trait introgression pipeline to allow for breeding with newly acquired herbicide traits.

   This report will not focus on the breeding methods as they are identical the methods in the nonGMO grant.

   Crossing: Sixteen unique glyphosate tolerant (GT) populations were initiated in the field in July 2023. This number is a large reduction from previous years as the GT project will not be continued in the future. However, these lines are promising for herbicide tolerance and Peking SCN resistance. One parent, ND17-26003(GT) was used in six of the populations due to its early maturity of MG 00.6. It is currently the earliest maturing line in the program but since it has the GT trait it can no longer be used as a parent. Parents were also selected for high yielding ability, IDC tolerance, and Peking SCN resistance. Six additional populations were sent to Costa Rica as F_2 seed. These populations were initiated in the greenhouse and have parents from the Illinois and Arkansas breeding program. The IL parents include: LD15-5776793, LD17-10157, and Arkansas line R01-581F (drought tolerance). F_1 seed was advanced to F_2 seed in a growth chamber at NDSU in summer 2023.

   Winter Nursery 2023/2024: F_1 and F_3 seed was shipped to the winter nursery at Costa Rica seeds in Upala, Costa Rica in November 2023. There were 50 F_1 seeds sent from 16 populations and two populations of GT F_3 seed sent. There was a failure of the crossing block in 2022 which accounts for the low population number of F_3 seed. F_1 seed will remain in Costa Rica for the entirety of early generation advancement. F_5 seed will be returned

from Costa Rica as single plant threshes in February/March of 2025. F_5 seed from the F_3 seed populations was shipped to North Dakota in spring 2024 for planting in North Dakota field season after two generations of advancement. F_2 seed from the Illinois and Arkansas populations will remain in Costa Rica until F_5 seed is produced. It will be returned in Fall 2024 or February/March 2025 depending on shipment dates and cost.

F_5 plant rows in 2023: In an effort to reduce unfavorable phenotypes such as dark brown pod, long internode, and thin stem, heavy selection against those traits was performed on F_5 plant rows. The glyphosate tolerant project is also being reduced as the program transitions to conventional germplasm with an herbicide trait introgression pipeline. Only 50 glyphosate tolerant plant rows are being tested in 2024 preliminary trials.

Preliminary yield trials 2023: 194 early, 158 mid, and 30 late maturing lines were tested in 2023 preliminary trials consisting of 2 rows/2 reps/2 locations in alpha lattice design. Maturity data was based on maturity notes taken during the F_5 plant row stage in 2022. Of these lines, 25 were selected to progress to second year yield trials based on yield and maturity alone.

Second year yield trials 2023: Second year yield trials were planted in four locations as 2 rows/ 2 reps. Sixty GT entries were trialed. All lines that enter second year yield trials are also phenotyped for *Phytophthora*, IDC, and SCN resistance. Based on the analysis of yield, maturity, and stacked disease resistances two lines were selected to advance to third year yield trials.

Advanced yield trials (3rd and 4th year) 2023: Since the GT pipeline will be ending, only lines that are eligible for variety release can be advanced to late stage testing two lines with SCN resistance and race 4 *Phytophthora* tolerance will be trialed in 4th year yield trials.

Variety Pre/Release- A GT line ND18-20092(GT) was accepted for prerelease in spring 2024. It is currently entered into another year of yield testing and IDC testing to ensure stability before it going into variety release in January 2025.

d. Research Results/Outcomes

Focus will be on the two lines advanced to 4th year yield trials and the prereleased ND18-20092(GT) line.

BLUPs were generated for planted maturity as days after Sept 1 and yield across site years. Additionally data for *Phytophthora* Race 4 tolerance, moderate resistance to full resistance to SCN HG type 2.5.7 and IDC tolerance could not be considered as the data from 2022 was not considered useful. A summary of the two lines entered into 4th year trials is found in Table 1.

Table 1. Summary of data for GT lines entered into 4th year yield trials					
Pedigree	Maturity after Sept 1.	Predicted MG	Yield	P. sojae tolerance	SCN
ND14-8899(GT SCN)*ND Bison(SCN)	16.73	0.4	48.5 bu/a	Race 4	Mod. Res- Full Resistance
ND14-8899(GT SCN)*ND11-19725(SCN)	14.8	0.1	45.04 bu/a	Race 4	Mod. Res- Full Resistance
Check 1 – AG0333	16	0.3	50.78 bu/a	Unknown	No SCN
Check 2- ND17009GT	13.2	00.9	41.93 bu/a	Race 4	No SCN
Check 3- ND21008GT20	11	00.8	41.38 bu/a	Race 4	No SCN

ND18-20092GT prerelease data

Maturity after Sept 1 data is shown in Table 2.

Table 2. 2021-2023 Maturity data for experimental line ND18-20092(GT) (in green) and checks												
PM Days after 9/1	MG	Predicted value	23_ ART	23_ CAS	23_ GRA	23_ LAM	23_ LIS	23_ MIL	22_ ART	22_ LIS	22_ MIL	21_ CAS
Variety		PM	PM	PM	PM	PM	PM	PM	PM	PM	PM	PM
AG005X8	00.5	9.5	5.3	7.0	2.3	8.0	8.5	7.0	7.0	7.0	8.3	2.3
AG0333	0.3	17.6	10.5	18.3	9.8	15.0	18.8	13.3	13.3	17.7	12.3	13.0
AG11X8	1.1	24.7	19.3	22.8	18.3	17.8	28.0	20.0	21.0	21.7	21.3	19.0
ND21008GT20	00.8	11.7	5.0	11.3	3.3	11.0	13.0	9.8	10.0	8.7	13.0	2.0
ND2108GT73	0.8	23.0	12.5	22.5	17.5	20.0	21.0	17.5	17.3	24.7	26.0	15.7
ND17009GT	00.9	13.4	5.0	13.3	4.0	10.0	11.8	9.5	9.3	14.7	13.3	.
ND18-20092(GT)	0.1	13.5	5.0	14.3	5.5	12.8	13.8	10.0	8.7	16.0	14.0	5.0

Yield data collected by NDSU breeding program in the Fargo area is shown in Table 3.

Maturity and yield data from research extension centers is shown in Table 4.

Table 3. 2021-2023 Yield for ND18-20092(GT) (in green) and checks								
	MG	Predicted value over years	23_ART	23_CAS	23_GRA	23_LAM	23_LIS	23_MIL
Variety		Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)
AG005X8	00.5	40.8	27.9	39.2	28.1	27.6	36.8	37.4
AG0333	0.3	50.2	40.9	49.0	35.8	44.8	53.2	44.1
AG11X8	1.1	52.7	42.2	57.6	41.6	48.8	53.2	42.9
ND21008GT20	00.8	40.6	25.1	38.2	29.7	31.3	38.1	40.3
ND2108GT73	0.8	48.9	37.0	52.3	37.5	46.4	55.1	51.6
ND17009GT	00.9	41.5	24.8	34.6	26.2	31.8	38.4	37.9
ND18-20092(GT)	0.1	41.6	29.6	41.7	26.4	31.1	47.1	47.6

Table 3 continued										
	MG	Predicted value over years	22_ART	22_CAS	22_LIS	22_MIL	21_ART	21_CAS	21_LIS	21_MIL
Variety		Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)
AG005X8	00.5	40.8	19.3	51.3	39.3	45.3	37.7	38.9	48.0	33.0
AG0333	0.3	50.2	40.2	63.9	43.0	63.9	47.9	40.4	48.9	46.2
AG11X8	1.1	52.7	40.4	66.6	59.5	62.2	47.4	27.9	60.4	47.0
ND21008GT20	00.8	40.6	27.6	55.1	35.1	52.4	34.5	13.1	43.3	32.3
ND2108GT73	0.8	48.9	16.0	59.0	53.7	43.1	34.8	50.8	58.5	46.8
ND17009GT	00.9	41.5	32.5	56.5	41.9	57.1	.	.	.	.
ND18-20092(GT)		41.6	24.0	54.3	38.0	48.1	26.8	38.8	51.7	35.2

Table 4. REC data			2022 Yield							
	Langdon	Park River	Langdon	Park River	Minot	Will Dry	Hettinger	Mandan	CAR Dry	CAR Ir
ariety	DTM	DTM	Yield	Yield	Yield	Yield	Yield	Yield	Yield	Yield
			bu/ac	bu/ac	bu/ac	bu/ac	bu/ac	bu/ac	bu/ac	bu/ac
G005X8	105.7	103.3	63.3	63.7	37.3	10.1	28.1	43.4	45.8	49.7
D21008GT20	111.7	107.3	60.6	65.2	30.3	11.6	24.2	43.3	48.4	58.0
D17009GT	113.7	110.3	55.0	58.5	31.7	9.5	22.9	35.9	44.2	43.2
D18-20092	116.7	110.7	57.8	64.1	33.2	9.5	24.4	38.4	47.1	50.8

IDC tolerance data is show in Table 5.

Table 5. 2023 IDC Score Data				
	Predicted values across locations	23_AME	23_COX	23_LEO
Variety	IDC	IDC	IDC	IDC
AG005X8	2.93	1.08	2.98	4.03
AG0333	3.20	1.65	3.15	4.50
AG11X8	3.29	1.73	3.03	4.95
ND21008GT20	3.14	2.15	2.90	3.98
ND2108GT73	3.37	2.45	3.15	4.45
ND17009GT	3.80	3.10	3.98	4.95
ND18-20092(GT)	3.30	1.90	3.75	4.08

SCN yield data in an infected field is show in Table 6.

Table 6. 2023 SCN Field Data (Yield)				
	Predicted value	23_SCN_ABS	23_SCN_COX	23_SCN_PRO
Variety	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)	Yield (bu/ac)
BARNES	24.24	11.10	32.89	26.62
MN0095	23.89	9.80	35.88	23.52
ND BENSON	32.85	11.51	47.44	46.29
ND BISON	27.54	10.35	41.77	31.74
ND15-22873(GT)	25.69	8.47	33.88	34.09
ND18-20092(GT)	31.53	18.52	44.74	36.64

SCN HG type testing was performed by Dr. Guiping Yan and is in Table 7.

Female Index (FI)= (mean number of females on test cultivar) ÷ (mean number of females on control) x 100

Rating scale

FI < 10	= Highly Resistant; HR
FI 10 to 24	= Resistant; R
FI 25 to 39	= Moderately Resistant; MR
FI 40 to 59	= Low Resistance; LR
FI > 60	= No Resistance; NR
FI > 25, CV > 35	= Not Determined; **

(*)=small root, (.)=missing sample, (**)=rep data too variable to rate

Table 7. Summary of HG type testing performed by Dr. Guiping Yan

Treatment	Obtained from	Planting date	Harvest date	Inoculum	# of females	Average # females	Female Index (%)	Resistance rating
Barnes R1	Ted Helms	2/10/23	3/14/23	HG 2.5.7	252	251		
Barnes R2	Ted Helms	2/10/23	3/14/23	HG 2.5.7	244			
Barnes R3	Ted Helms	2/10/23	3/14/23	HG 2.5.7	194			
Barnes R4	Ted Helms	2/10/23	3/14/23	HG 2.5.7	312			
ND18-20092 (GT) R1	NDSU Breeding Program	2/10/23	3/14/23	HG 2.5.7	60	67	26.5	MR
ND18-20092 (GT) R2	NDSU Breeding Program	2/10/23	3/14/23	HG 2.5.7	68			
ND18-20092 (GT) R3	NDSU Breeding Program	2/10/23	3/14/23	HG 2.5.7	60			
ND18-20092 (GT) R4	NDSU Breeding Program	2/10/23	3/14/23	HG 2.5.7	78			

e. Disclosure of variety release

ND18-20092(GT) was accepted for prerelease and is eligible for consideration for variety release in January 2025 as a GT tolerant SCN resistant variety with moderate resistance to HG type 2.5.7.

f. Discussion:

The conversion of the glyphosate tolerant project to a trait introgression pipeline is a welcome modernization change to the program. A major success of the 2023/2024 funding year was the acquisition of the BASF Liberty trait. Seed was delivered in May 2024. The maturity is MG VI which will be slightly challenging but fortunately we have a growth chamber setup to facilitate crossing across maturity groups. The first cross is expected in July 2024.

Applying heavy selection pressure to the all GT lines to facilitate advancement of only variety release candidates instead of parents has made managing this project much easier.

The potential variety release of ND18-20092(GT) is a milestone to look forward to as it will be the first GT and SCN resistant line the breeding program has released. The yield results in SCN infected fields makes this line very competitive.

g. Conclusion:

The glyphosate project has served ND farmers well and has release very successful lines such as ND17009GT which at one point was grown on 2.4% of all soybean acres in the state. However with the demand for newer herbicide resistances due to improved technology and evolving weed resistance to glyphosate, it is appropriate to listen to the market and introgress new herbicide traits. With the acquisition of the BASF Liberty trait, and the creation of new populations utilizing high yield lines from other maturity groups, plus to focus on breeding for Peking style SCN resistance and IDC tolerance, there is optimism that the NDSU soybean breeding program will produce new varieties that will be competitive throughout the state. There will be a lull in variety release while this happens, but the expedited pipeline that was created taking advantage the growth chambers in the NDSU greenhouse and faster growing season of equatorial Costa Rica is ensure new materials are ready to test in only a few years. North Dakota farmers will be growing improved, high yielding NDSU soybean varieties before they know it.