



# DEVELOPMENT OF LONG-TYPE CONFECTION SUNFLOWER HYBRIDS

YEAR 5: 2022-2023 Performance Report

March 31, 2023



## **ACTIVITY 1: SUMMER AND WINTER NURSERY**

The objective of the summer and winter breeding nurseries is to develop elite parent lines, possessing genes for tolerance to sulfonylurea herbicides, rust and downy mildew that will, when crossed, produce herbicide tolerant experimental hybrids highly adapted to Canada, with a prominent level of resistance to downy mildew and rust, and possess improved seed types for Canadian processors and producers. For simplicity, the breeding of male and female parent lines in our program generally follows the same process.

The 2022 summer nursery was planted near Fargo, North Dakota with a total of 3,000 rows.

The goals for the 2022 nursery included:

- Generation of new breeding populations (male restorer and female AxB lines) with desired characteristics from crossed plants.
- Selection in segregating populations for yield, seed type, agronomic integrity, and presence of genes for herbicide tolerance and disease resistance.
- Select early and late generation populations at contra season nursery.
- Identification of new homozygous parent lines resistant to rust and downy mildew for use in production of preliminary hybrids.
- Identification of genetic material with the herbicide tolerance gene.
- Advancement of male restorer lines and female parent lines to semi-elite or elite category.
- Interim report on agronomic ratings and disease resistance.

Activity 1 met its Year 5 (2022-23) objectives to develop elite parent lines, possessing genes for tolerance to sulfonylurea herbicide (SU-7: non-transgenic), rust ( $R_{12}$ ) and downy mildew ( $PL_{ARG}$ ) to provide a competitive production advantage to existing hybrids. The following quantities of finished, and unfinished parent lines were grown. The quantities of lines contained in the program are as follows:

- Finished male restorer lines no dominant disease resistance genes. Elite (19)
  - Due to an increased emphasis to develop lines that contain dominate genes for disease resistance, no additional elite lines were added in 2022 that do not contain disease resistance
- Finished male restorer lines no dominant disease resistance genes. Semi- Elite (11)
  - Due to a desire to shift away from male parent lines that do not contain genes for disease resistance. No semi elite male lines were added that contain no genes for disease resistance.
- Finished male restorer lines fixed for gene PLARG or PLARG and R12 Elite (45)
  - Several male lines containing disease resistance genes were used for the third year in hybrid combinations in 2021. 10 lines were advanced to elite based on hybrid performance.
- Finished male restorer lines fixed for gene PL<sub>ARG</sub> or fixed for both PL<sub>ARG</sub> and R<sub>12</sub>. Semi-Elite (67)
  - Additional male lines containing disease resistance genes were used for the second year in hybrid combinations in 2021. 16 lines were advanced to semi elite based on hybrid performance.



- Finished female A x B lines with cytoplasmic male sterile conversion completed. Elite (26)
  - 3 additional finished A x B lines were used for a third year in hybrid combinations in
     2021. The lines were advanced to elite based on hybrid performance.
- Finished female A x B lines with cytoplasmic male sterile conversion completed. Semi-Elite (47)
  - 2 additional finished A x B lines were used for the second year in hybrid combinations in 2021.
- Based on the 2020, 2021 and 2022 SNP marker data, there currently are approximately 322 male lines (finished + unfinished) in the program fixed for the downy mildew resistance (PLARG).
  - 205 lines were added, and 60 previous sub-lines were discarded due to redundancy and/or re-evaluation.
- Based on the 2021 SNP marker data, several lines that had previously tested positive for the rust gene R12 no longer tested positive. It was determined that NSA marker 001570 C2 was not proving to be as determinate as previously thought for gene R12. NSA marker 005061 has been added to the protocol, appears to be highly dependable and will be used as our premier SNP marker for R12 screening. The MCA program also contains a rust gene R13. R13 is more effective than R12 for resistance to rust. R13 does not have an SNP marker and is tracked in the program by observing rust resistance of lines and hybrids in the field. Currently there are approximately 151 Male lines (finished + unfinished) in the program fixed for both downy mildew resistance (PLARG) and rust resistance either(R12) or (R13)
- A new female development program that includes PL<sub>ARG</sub> and R<sub>12</sub> was initiated in the 2017-18 winter nursery. Based on the 2022 SNP marker data, there are 11 finished female A x B lines containing PL<sub>ARG</sub> (downy mildew) or contain both PL<sub>ARG</sub> and R<sub>12</sub>, (rust). Due to the problem encountered with NSA marker 001570 C2 and the transitioning over to NSA marker 00561 C2, a new female development program that includes PL<sub>ARG</sub> and R<sub>12</sub> was re initiated in the 2021-22 winter nursery, using lines fixed for rust marker 005061 C2. Having PL<sub>ARG</sub> and R<sub>12</sub> on both sides of the program (male and female) will increase the opportunity for resistance to downy mildew and rust in experimental hybrids in cases where the male line may lack genes for resistance.

# Generation of new breeding populations (male restorer and female AxB lines) with desired characteristics from crossed plants

#### **Development of Male Restorer Lines:**

#### **SUMMER 2022**

• Plants were selected from F<sub>2</sub> populations. Selected F<sub>2</sub> plants were rated for agronomics and seed type.



- Tissue samples were taken from the top rated F<sub>2</sub> plants and marker screened for dominant disease resistance genes.
- F<sub>4</sub> plants were selected. Selected F<sub>4</sub> plants were rated for agronomics and seed type.
- Tissue samples were taken from the top rated F<sub>4</sub> plants and marker screened for dominant disease resistance genes.

#### **WINTER 2022-23**

- F<sub>3</sub> plants were selected in the winter nursery (Chile) based primarily on earliness and plant type.
- F<sub>5</sub> plants were selected in the winter nursery based primarily on earliness and plant type.

#### Development of Female AxB Lines:

#### **SUMMER 2022**

- F<sub>2</sub> plants were selected before bloom based on earliness and plant type and a first cross was made to cytoplasmic male sterility.
- The top rated F<sub>2</sub> plants (AxB pairs) were advanced to winter nursery based on agronomics and seed type.
- F<sub>4</sub> plants (AxB pair) were selected based on earliness and plant type with a third backcross made to cytoplasmic male sterility.
- The top rated F<sub>4</sub> plants (AxB pairs) were advanced to the winter nursery based on agronomics and seed type.

#### **WINTER 2022-23**

- F<sub>3</sub> plants (AxB pairs) were selected based on earliness and plant type and a second backcross was made to cytoplasmic male sterility.
- F₅ plants (AxB pairs) were selected based on earliness and plant type with a fourth backcross made to cytoplasmic male sterility. If female conversion to sterility was complete, the new female A x B line was used to make experimental hybrids for testing in the 2021 summer nursery.



Figure 1. Chile winter nursery.





#### **RESULTS**

- Selection in segregating populations for yield, seed type, agronomic integrity, and presence of genes for herbicide tolerance and disease resistance
- Identified new homozygous parent lines for production of preliminary hybrids.
- Screened new parental line material for resistance to rust (R<sub>12</sub> gene), downy mildew (PL<sub>ARG</sub>).
- Screened material for the herbicide tolerance gene SU-7.
- Collected agronomic ratings on maturity, lodging, height, disease, and a general visual screening of materials.

# **Herbicide Tolerance Screening**

All parent lines in the program were developed with herbicide tolerance. The SU-7 gene is a single dominant gene from DuPont that conveys herbicide tolerance to tribenuron in sunflower. The trait has been incorporated into 100% of the program parental lines and early generation material, after successful completion of the five-year funding under GF2 "Confection Sunflower Development Initiative".

Both the summer and winter nurseries are sprayed with a two times rate of tribenuron to confirm that the presence of the trait within the hybrids. Any hybrids that show injury are discarded.

MCA collected tissue samples in the summer of 2022 to confirm the prevalent races found in Manitoba.

# **Downy Mildew, Rust, and Sclerotinia Screening**

Breeding activities in the male and female parent line program include the incorporation of genes for disease resistance (downy mildew: PL<sub>ARG</sub>, rust: R<sub>12</sub>).

Male and female parent lines that contain the resistant genes were screened to confirm that the resistance genes were present. Plants that contain the resistance genes were selected for further advancement into the 2022-23 winter nursery.

Fifty-four new herbicide tolerant male restorer lines ( $F_5$ ) that are fixed for downy mildew resistance were identified. All fifty-four lines are fixed for  $PL_{ARG}$ , or  $PL_{ARG}$  and either R12 or R13. An additional 19 new lines were identified and used in the winter breeding nursery to make experimental hybrids for testing in 2022 and were thus advanced to activity 2. 9 of these lines are fixed for  $PL_{ARG}$ , or  $PL_{ARG}$  and either R12 or R13.

Based on agronomics, seed type and SNP marker information obtained from individual plants, 10 new F5 female lines were identified that contain genes for downy mildew and/or rust resistance and were advanced to the 2022-23 winter nursery. Conversion to cytoplasmic male sterility (CMS) of the 10 new female lines was initiated in the 2018-19 winter nursery. CMS conversions were completed in the 2021 summer nursery. The 10 new female lines containing genes for resistance were advanced to the 2022-23 winter nursery and will be used to make experimental hybrids for testing in Manitoba in summer 2023.



Currently in the program, there are approximately 266 finished parent lines that have been utilized to make experimental hybrids used for testing in Canada (190 males, 76 females) since the program started testing herbicide tolerant hybrids in 2014. All 266 lines are herbicide tolerant and have been advanced through the program based on solid agronomics, seed type and adaptability to Canadian growing conditions. 148 of the male lines contain genes for resistance to downy mildew, or downy mildew and rust. 14 of the female AxB lines contain genes for resistance to downy mildew, or downy mildew and rust.

Based on comparative yields and seed types obtained from experimental hybrids tested in Manitoba in 2022, it appears that the program currently has the potential to produce a high number of experimental hybrids that can yield competitively and can produce improved seed types to commercial confection hybrids currently being grown in Canada. High yield performance and improved seed types in combination with herbicide tolerance and genetic disease resistance will provide attractive hybrid options for Canadian producers.





## **ACTIVITY 2: CANADIAN TESTING PROGRAM**

The overall objective of the Canadian Testing Program is to isolate commercially viable experimental hybrids for advanced testing and eventual commercialization. While seed type and marketability are of extreme importance, the hybrids must also be early maturing, high yielding and have a strong agronomic package. Testing activities will include four levels of testing.

# **Preliminary Hybrid Screening**

Planting Date: May 27 (Elm Creek) and May 27 (Holland)

The MCA transports our planting equipment from Fargo, ND to plant the two preliminary screening nurseries. This process has eliminated planting errors and ensures that the trials are planted at the desired time.

132 new preliminary hybrids were selected from the 2020-2021 Summer and Winter nurseries to be

tested at two locations in Manitoba. Each location consisted of two row plots that were replicated twice. The trials included two commercial performance checks (6946 DMR and Panther DMR) and one SU-7 herbicide tolerant check (P63ME70). The two commercial performance checks are not tolerant to the SU-7 herbicide, so they were strategically placed along the edges of the trials. The nursery was sprayed with conventional herbicides and hand weeded if needed.



Figure 2. Holland, MB Sunflower Hybrid Nursery

The two Manitoba nursery locations included an "Advance Yield Trial" (AYT) that had a selection of elite hybrids selected from the Preliminary trials in 2021. 18 elite hybrids were selected for a second year of testing alongside the preliminary nursery. The AYT had two row plots that were replicated three times. Data collected from both the preliminary screening nurseries and the AYT included standard agronomic traits for: emergence, herbicide tolerance, days to bloom, days to maturity, plant lodging, seed yield, seed test weight, seed sizing and percentage of nut-meat ratio.

The trial was sprayed and tilled by a contract services company in both locations and plot maintenance including thinning was performed by MCA's Research Trial Specialist with assistance from MCA staff. Benchmarks for a commercial confectionary hybrid were:





- · Herbicide tolerance: resistance to sulfonylurea herbicide: SU-7;
- · Seed Type: Dark color, long (1.9-3.2 cm) seed with shoulder width;
- · Disease resistance: resistance to Downy Mildew (PL<sub>arg</sub>) and Rust (R<sub>12</sub>);
- · Early maturity: less than or equal to the commercially available hybrid check or about 117 days to R9 maturity;
- · Improved yield over commercially available hybrids;
- · General plant integrity/agronomics acceptable height, good lodging tolerance, and good overall agronomic package.

MCA's Research Trial Specialist and contract breeder hand clipped and threshed the sunflower heads from the selected lines on October 11th (Holland) and October 11<sup>th</sup> and 12<sup>th</sup> (Elm Creek, MB). The harvested sunflower seed was dried before samples could be weighed. The seed from the selected plots were cleaned, weighted for yield and bushel weights were calculated.

Of the 132 new preliminary lines evaluated in 2022, 52 were taken to harvest and evaluated for yield, agronomic and seed quality characteristics. Based on the overall performance along with seed appearance and quality, 18 hybrids have been selected for the next level of testing in 2023. Seed of the selected 18 hybrids is being multiplied in the 2022/23 winter nursery in Chile for trialing in Manitoba in 2023.

# **Variety Performance Trials**

To evaluate the elite hybrids against other commercially available sunflower hybrids for their agronomic competitiveness, merit, and regional adaptation. Each Variety Performance Trial consisted of three replications using a RCBD (Randomized Complete Block Design) for data analysis.

Five elite hybrids were tested in the VPTs at 4 locations in Manitoba in 2022. The locations were: Melita, Carberry, Rossendale, and the Elm Creek/Carman area. All 4 locations were planted and taken to harvest. The Melita trial was lost due to drought conditions that persisted at the location from the middle of July to the middle of September. The trials located at Rossendale and Elm Creek area were planted late in the spring due to wet field conditions but still managed to reach maturity. The complete set of trial results were made available on both the MCA and Seed Manitoba websites in the winter of 2022. The MCA website link: Manitoba Crop Alliance | Sunflower Variety Performance Trials (mbcropalliance.ca).





**Table 1: 2022 Variety Performance Trial Results** 

SUNFLOWERS - C	ONFECTIONARY TYP	E							
Comments:									
These varieties w	ere tested and data	donated by the	Manitoba C	rop Allianc	e (MCA).				
	rieties listed are sus								
Genetic resistano	e to verticillium wilt	is rated as mod	lerately susce	eptible to n	noderately res	sistant for all s	unflower vai	ieties presen	ted.
Plant population	and environment w	ill contribute gr	eatly to the f	inal produc	t.				
Variety Description	ons								
		Genetic	Site	Yield	Maturity <sup>2</sup>	Height	Seed Sizing (%)		3
Company	Hybrid	Traits <sup>1</sup>	Years	% Check	(+/- check)	(+/- check)	>22/64	>20/64	<20/64
NuSeed	6946 DMR	DM	34	100	0	0	20	39	41
NuSeed	Panther DMR	DM	42	99	0	-2	41	31	28
	es tested/proposed			33				01	20
CHS Sunflower	20-EXP3	ExSun	6	95	5	0	58	28	14
CHS Sunflower	21-EXP1	ExSun	6	91	7	2	37	36	27
MCA	EX 20057	ExSun	3	93	1	1	57	22	21
MCA	EX 20306	ExSun	3	87	0	-2	71	17	12
MCA	EX 200239	ExSun	3	84	1	2	77	14	9
MCA	EX 359239	ExSun	3	85	2	4	79	12	10
MCA	EX 570309	ExSun	3	92	-2	2	48	22	30
NuSeed	NJKE05926	ExSun	3	92	-1	-2	4	13	83
NuSeed	NJKM65823	CL	3	84	6	0	28	39	33
NuSeed	NDKM15700	CL	3	94	2	-5	46	29	25
NuSeed	NDKM16761	CL	3	88	4	-3	35	35	30
	CHECK CHARACTERISTICS								
	6946 DMR		34	2977	121	64			
			site years	lb/ac	days	inches			
1 Genetic traits in	clude CL = Clearfield	tolerance; ExS	un = Express	tolerance;	DM = Downy	Mildew Resis	tance.		
2 Physiological m	aturity for sunflowe	rs is R9, where	the bracts or	the head a	are almost co	mpletely brow	/n.		
3 Totals may not	add to 100% due to	rounding; infor	mation base	d off single	year data for	three sites at	Carberry, Eli	m Creek, and	Rossenda
Refer to the MCA	website at www.ml	ocropalliance.c	a for more de	etails.					

# **Commercial Strip Trial Testing**

Advanced successful experimental hybrid(s) to the next level of testing, pre-commercial strip trial testing.

Five elite hybrids were selected from the 2021 Preliminary Nursery and were proposed for testing at 3 locations in a field scale strip trial comparison with a commercially available hybrid. The field trials were planned to be a minimum of 8 seeded rows each by a minimum length of 1000'. The elite hybrids in the strip trial program are to be tested in the same manner as the producers commercially available hybrid. The elite hybrids would be tested within the producer's commercial field, with their own equipment using their production practises.

Three producers were lined up for planting the head-to-head strip trials, but due to the late, wet spring season the three cooperating producers adjusted their planting intentions. One producer changed crop types due to the late season, and two moved from confection to oilseed sunflowers. As a result of the delayed planting season, confection-type sunflower planting acreage in Manitoba reduced from 15,652





acres in 2021 to 4,492 acres in 2022.

# **Commercial Pilot Scale Testing**

Seed production for testing elite hybrids under a pilot scale in comparison to a commercially available hybrid.

Select hybrids would have seed production conducted in the winter nursery located in Chile. Elite hybrids would be grown on a small acreage in comparison to a commercial hybrid. The pilot scale testing will be done using a farmer's equipment using their best management practices for sunflower production.

Testing of the advanced hybrids had only been to the pre-commercial strip trial testing stage. In the fall of 2022, 3 of the 5 elite hybrids were selected for pilot scale testing in the 2023 growing season by a commercial seed processor. Seed production of these 3 hybrids was initiated for winter seed increases in cages in Chile in the fall of 2022.



Figure 3. Three pre-commercial hybrids: EX2002239; EX359239; EX20306.





#### **KNOWLEDGE TRANSFER EVENTS**

As the producer organization for Manitoba sunflower growers, knowledge transfer is extremely important, MCA must demonstrate producer's check-off dollars at work. For this project, there are two specific target audiences: producers and sunflower processors/buyers.

For 2022-2023, Manitoba Crop Alliance continued to adjust our knowledge transfer events due to COVID-19. Year 5 progress on the Development of Long-Type Confection Sunflower Hybrids project was communicated with members and industry representatives through various methods:

- Seasonal tweets throughout the growing season
   Twitter: @mbcropalliance
   Heads Up newsletter
  - Fall 2022 Processor Tour
- · Fall 2022—Walk the Plots video: https://www.youtube.com/watch?v=Qphhnb5hAnQ.



Figure 5. MCA social media knowledge transfer.

Figure 4. Processor/Industry Tour, Elm Creek, MB.

