

Transfer of Sclerotinia Basal Stalk Resistance from Wild Annual *Helianthus* Species into Cultivated Sunflower (*Helianthus annuus* L.)

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Abstract

Sclerotinia basal stalk rot (BSR) is caused by *Sclerotinia sclerotiorum*, a necrotrophic fungus capable of causing a stem disease that is a major yield-limiting factor in global sunflower production. Sunflower is the only host species that is susceptible to BSR as a root infection. BSR resistance breeding is relatively complex, since resistance is controlled by multiple genes each with a small effect. To increase the level of resistance and genetic diversity of cultivated sunflower, wild annual *Helianthus* species were evaluated for resistance to Sclerotinia BSR with four annual species showing high levels of tolerance which were introgressed into cultivated sunflower. Interspecific hybrids were screened in artificially inoculated field trials over six environments. Eight interspecific germplasm lines based on four annual species were identified: CMS 89 x PAR-1673 (Texas); CMS 451 x PRA-HIR 437 (Texas); CMS 441 x ANN-1 (Texas); CMS 441 x ANN 412-2 (Texas); CMS 466 x ANN 14 (Aust.); CMS 451 x ARG-130-1 (Mozb.); CMS 467 x ARG 130-2 (Mozb.); and CMS 451 x ARG 1 (S. Afr.). Average disease incidence (DI = percent of plants infected) for CMS 89 x PAR-1673 (Texas) was 11%; CMS 451 x PRA-HIR 437 (Texas) (16%); CMS 441 x ANN-1 (Texas) (5%); CMS 441 x ANN 412-2 (Texas) (6%); CMS 466 x ANN 14 (Aust.) (8%); CMS 451 x ARG-130-1 (Mozb.) (11%); CMS 467 x ARG 130-2 (Mozb.) (4%); and CMS 451 x ARG 1 (S. Afr.) (5%). The susceptible hybrid checks Cargill®270 had a DI of 60% and Hybrid 894 (47%), while the tolerant hybrid check Croplan®305 had 20%. The recurrent parents, CMS HA 89 was 36%, CMS 441 (20%), CMS 451 (44%), CMS 466 (19%), and CMS 467 (16%). The germplasm lines can be used in sunflower breeding programs to introgress Sclerotinia BSR resistance genes into other adapted lines providing a more efficient, durable, and environmentally friendly host plant resistance.

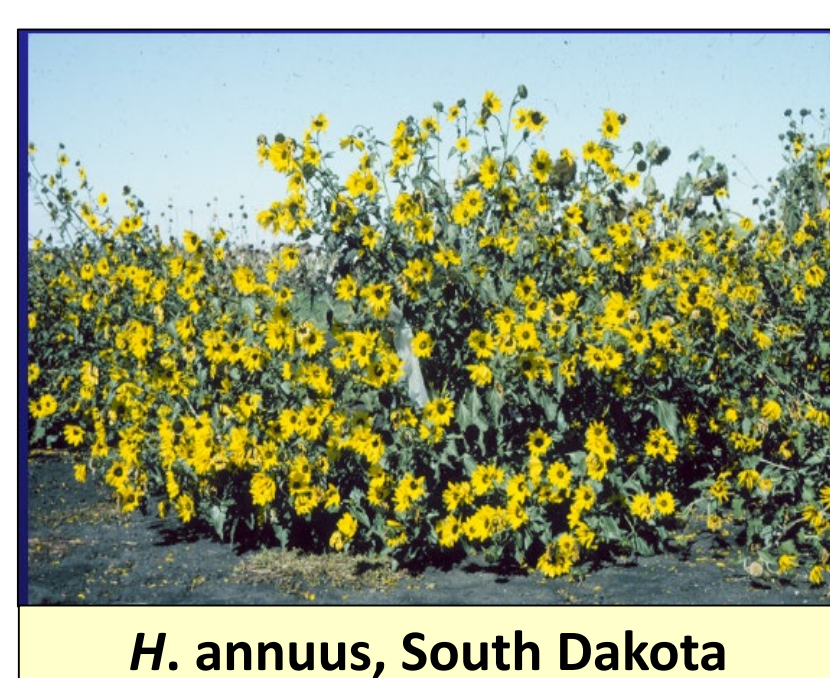
Introduction

Sclerotinia fungal diseases are considered the most important biological yield-limiting factors for sunflower production. The necrotrophic fungus *Sclerotinia sclerotiorum* (Lib.) de Bary (commonly called 'white mold' or Sclerotinia wilt) is among the most common and most devastating diseases, causing a significant threat to worldwide sunflower in humid temperate, as well as, tropical and sub-tropical regions of the world, causing economic losses by reducing seed yield and quality (Markell et al., 2015). The disease cycle of Sclerotinia basal stalk rot (BSR) begins with a root infection from mycelia of germinating sclerotia (Gulya et al., 2008). The management tools for Sclerotinia are insufficient: crop rotation is of marginal use due to the long-lived nature of the sclerotia, foliar fungicide application (commonly used for management of white mold in other crops) is not useful due to the unique myceliogenic infection process in sunflower, fungicide seed treatments provide insufficient control, and the present-day hybrids and cultivated lines lack sufficient tolerance and resistance (Seiler et al., 2017). Breeding using resistant germplasm is the most economic and efficient disease management strategy for most sunflower breeding programs. BSR is controlled by multiple genes each with a small effect with no major gene(s) identified. Significant variation in resistance has been reported in the annual sunflower crop relatives in the primary and secondary genes pool of sunflower (Block et al., 2012; Seiler et al., 2017). The objective of this breeding effort was to develop highly tolerant Sclerotinia BSR sunflower germplasms by transferring resistance from wild annual crop relatives into cultivated sunflower.

Materials and Methods

Introgression of Sclerotinia resistance from annual crop wild relatives

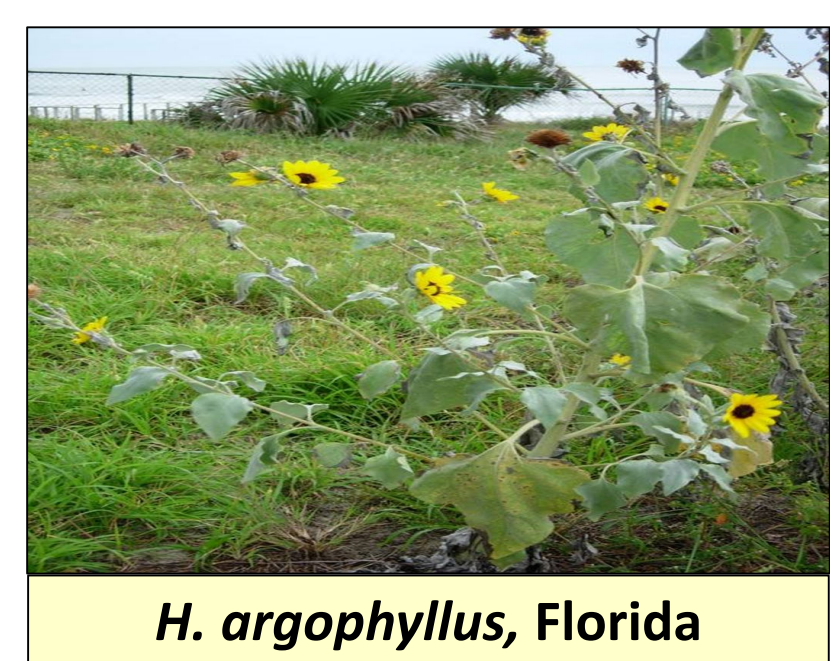
Annual Species



H. annuus, South Dakota



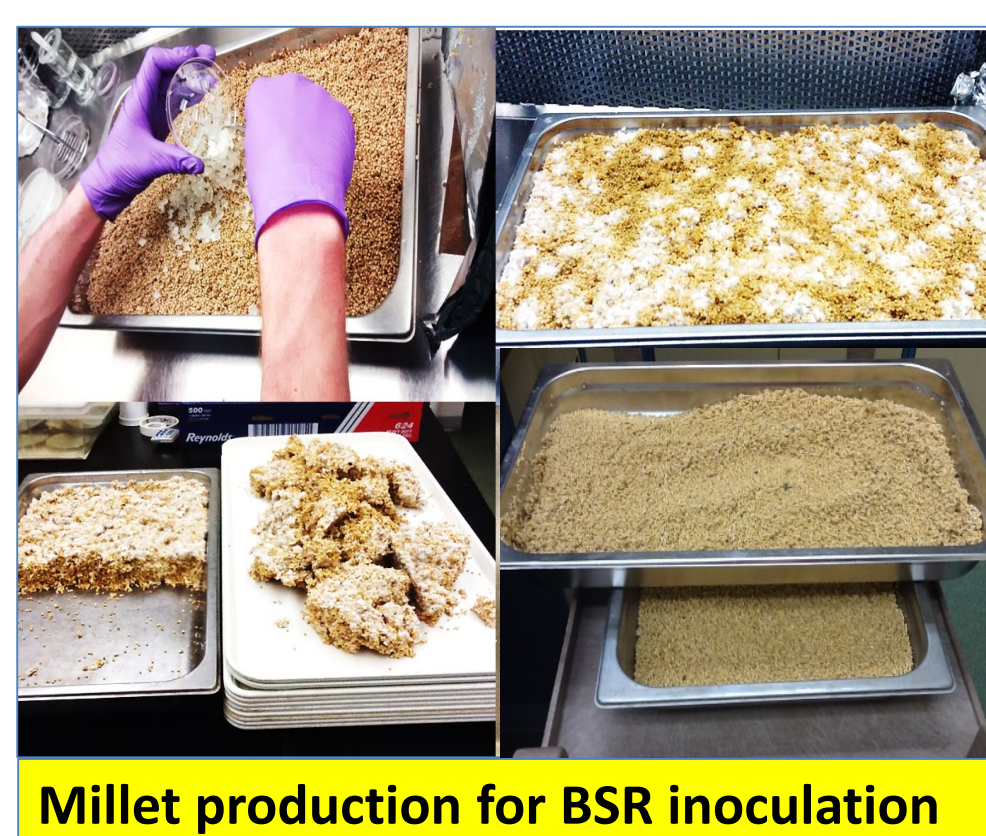
H. praecox ssp. *hirtus*, Texas



H. argophyllus, Florida



H. paradoxus, New Mexico



Millet production for BSR inoculation



Field inoculator for basal stalk rot



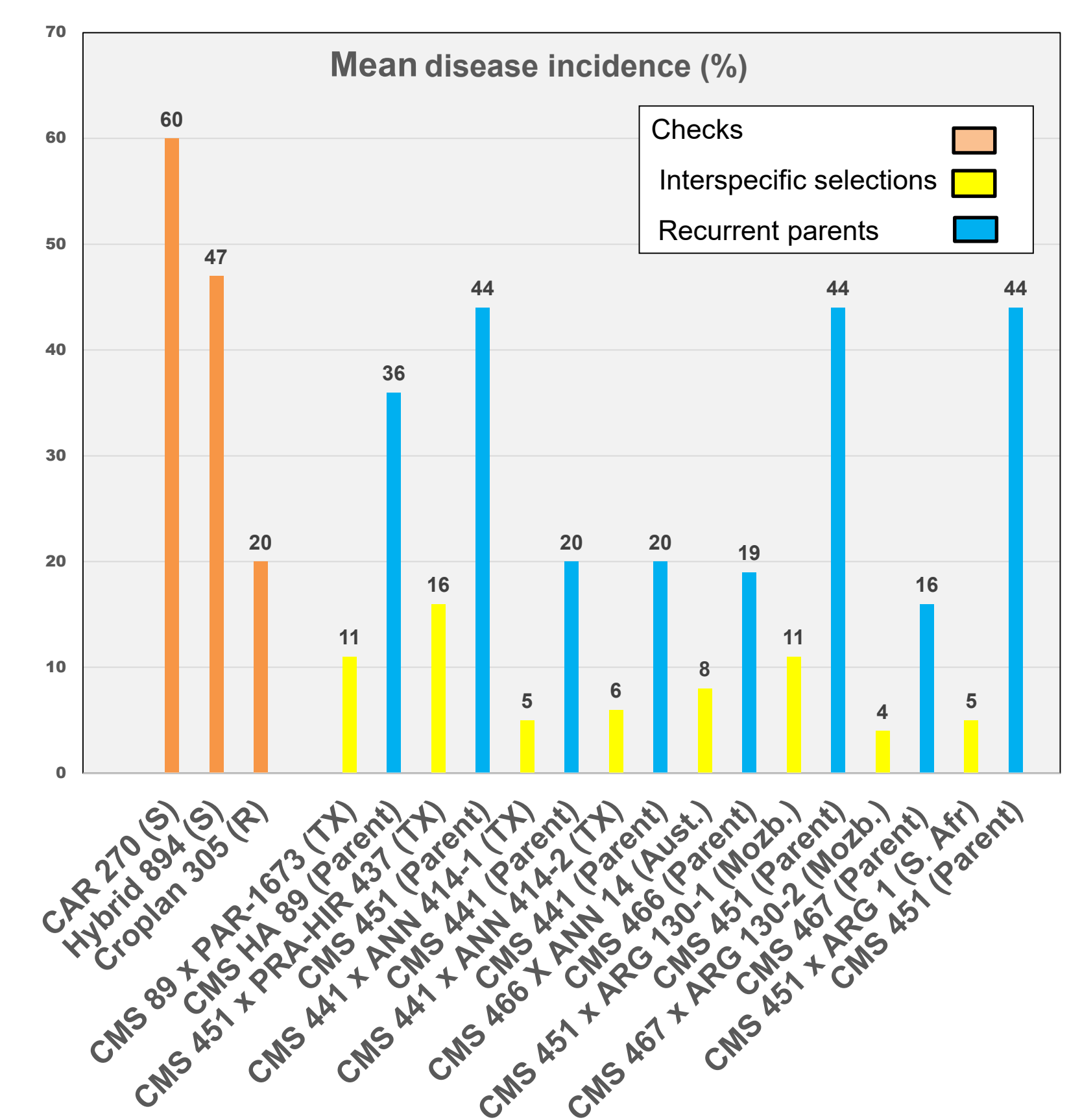
Sclerotinia stalk rot symptoms

Results and Discussion

Table 1. Basal stalk rot resistant germplasms based on field screening from 2014-2019

Line	Species	Mean DI (%)
CHECKS		
CAR® 270 (Susceptible)	Hybrid	60
Hybrid 894 (Susceptible)	Hybrid	47
Croplan® 305 (Resistant)	Hybrid	20
PARENTAL LINES		
CMS HA 89 (Parent)	Inbred Line	36
CMS 441 (Parent)	Inbred Line	20
CMS 451 (Parent)	Inbred Line	44
CMS 466 (Parent)	Inbred Line	19
CMS 467 (Parent)	Inbred Line	16
BREEDING LINES		SPECIES
CMS 89 x PAR-1673 (TX)	<i>H. paradoxus</i>	11
CMS 451 x PRA-HIR 437 (TX)	<i>H. praecox</i> ssp. <i>hirtus</i>	16
CMS 441 x ANN 414-1 (TX)	<i>H. annuus</i>	5
CMS 441 x ANN 414-2 (TX)	<i>H. annuus</i>	6
CMS 466 X ANN 14 (Aust.)	<i>H. annuus</i>	8
CMS 451 x ARG 130-1 (Mozb.)	<i>H. argophyllus</i>	11
CMS 467 x ARG 130-2 (Mozb.)	<i>H. argophyllus</i>	4
CMS 451 x ARG 1 (S. Afr.)	<i>H. argophyllus</i>	5

Figure 1. Mean disease incidence of eight BC₅F₂ families from wild annual species for Sclerotinia stalk rot resistance



Interspecific germplasms based on annual crop wild relatives of sunflower, *H. annuus*, *H. praecox* ssp. *hirtus*, *H. argophyllus*, and *H. paradoxus* were field screened in replicated BSR rain-fed field trials from 2014-2019. Increased tolerance to BSR was observed confirming the successful gene introgression of Sclerotinia BSR tolerance from several the annual wild species into cultivated sunflower. Field screening of interspecific germplasm for BSR at six different trials in North Dakota and Minnesota indicated high levels of tolerance for eight germplasm lines compared to the recurrent parent and hybrid checks (Table 1 and Figure 1). Across environments, the selected introgressed germplasms showed significantly lower Sclerotinia BSR DI than both susceptible hybrid checks, Cargill 270® (60%), Hybrid 894 (47%), and the resistant check hybrid Croplan 305® (20%), and parental CMS HA 89 (36%), CMS 441 (20%), CMS 451 (44%), CMS 466 (19%), and CMS 467 (16%). The lowest DI was observed in interspecific germplasm lines CMS 467 x ARG 130-2 (Mozb.) (4%), CMS 441 x ANN 414-1 (TX) (5%), CMS 451 x ARG 1 (S. Afr.) (5%), and CMS 441 x ANN 414-2 (TX) (6%).

Summary

Replicated field evaluations for BSR DI from 2014-2019 confirmed the successful introgression of Sclerotinia BSR tolerance from annual crop wild relatives resulting in germplasm for sunflower breeding programs.

References

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- Block, C., T.J. Gulya, and L.F. Marek. 2012. Identifying resistance to Sclerotinia stalk and root rot in perennial germplasm. In: Proceedings of the APS Annual Meeting, Providence, RI. August 4-8, 2012. American Phytopathological Society Press, St. Paul, MN. 269. http://www.apsnet.org/meetings/Documents/2012_Meeting_Abstracts/aps12abP269.htm.
- Markell, S.G., R.H. Harveson, C.C. Block, and T.J. Gulya. 2015. Sunflower diseases. In: E. M. Force, N. T. Dunford, and J. L. Salas, editors, Sunflower: chemistry, production, processing and utilization. American Oilseed Chemists Society Press, Urbana, IL. p. 129-156.
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Breeding Lines BC₂F₅

CMS 89 x PAR-1673 (Texas)
 CMS 451 x PRA-HIR 437 (Texas)
 CMS 441 x ANN 414-1 (Texas)
 CMS 441 x ANN 414-2 (Texas)
 CMS 466 X ANN 14 (Australia)
 CMS 451 x ARG 130-1 (Mozambique)
 CMS 467 x ARG 130-2 (Mozambique)
 CMS 451 x ARG 1 (South Africa)

Field Evaluation

- Environments: Carrington, ND, 2014-2015, 2017-2019; Staples, MN, 2019
- Design: Complete randomized design with 2 reps
- Inoculation: *S. sclerotiorum* mycelia grown on millet and deposited (90 gm) in furrows next to the rows of sunflower lines at V-6 growth stage
- Checks: CAR 270 (Susceptible), Hybrid 894 (Susceptible), and CROPLAN 305 (Resistant)
- Disease scoring: Disease incidence (DI), percent of plants showing disease symptom