

Evaluation and validation of sunflower genotypes with resistance to *Sclerotinia* basal stalk rot.

Angidi Srushtideep¹, Julie S. Pasche¹ and William Underwood².

¹ Dept of Plant Pathology, North Dakota State University, Fargo, ND, USA.

² USDA-ARS, Sunflower & Plant Biology Research Unit, Fargo, ND, USA.

Introduction

Basal stalk rot (BSR) of cultivated sunflower (*Helianthus annuus* L.) caused by necrotrophic fungal pathogen *Sclerotinia sclerotiorum* (Lib) de Bary is an economically significant disease limiting sunflower production in the Northern Great Plains region of the USA. Infection leading to BSR begins in sunflower roots and fungus subsequently move into the base of stem causing basal stem lesions, plant wilting, and premature senescence or death resulting in yield loss. Resistance to *S. sclerotiorum* is quantitative and no major genes having a large influence on the level of resistance have been discovered (1). Consequently, the improvement of host plant resistance is an important goal for sunflower breeding because producers have limited options for managing this disease. Therefore, the objectives of the study are

1. Distinguish between highly and moderately resistant genotypes by re-evaluating 60 cultivated sunflower genotypes that have shown some degree of resistance in field trials using an improved phenotyping method.
2. Evaluate selected genotypes with diverse *S. sclerotiorum* isolates to confirm broad-spectrum resistance.
3. Evaluate genotype X isolate interactions.

Methodology

- The greenhouse phenotyping experiment was conducted three times, resulting in days to death data for 48 plants per accession.
- After inoculation (Figure 2), plants were evaluated for 28 days, the number of days was recorded when each plant exhibited terminal wilt or whole- plant desiccation (Figure 3).
- Levene's test was performed to confirm homogeneity of variance and the combined experiments were later analyzed by a generalized linear model. Tukey's post-hoc test was used to identify significant differences among the lines



Figure 2: Inoculation of Sunflower genotype

A. Five-week-old sunflower genotype. B. Half teaspoon (0.38g) millet inoculum. C. Five-week-old root bound plant removed from sheet pot. D. Millet inoculum placed at the bottom of the sheet pot. E. Plant placed back in inoculated sheet pot.

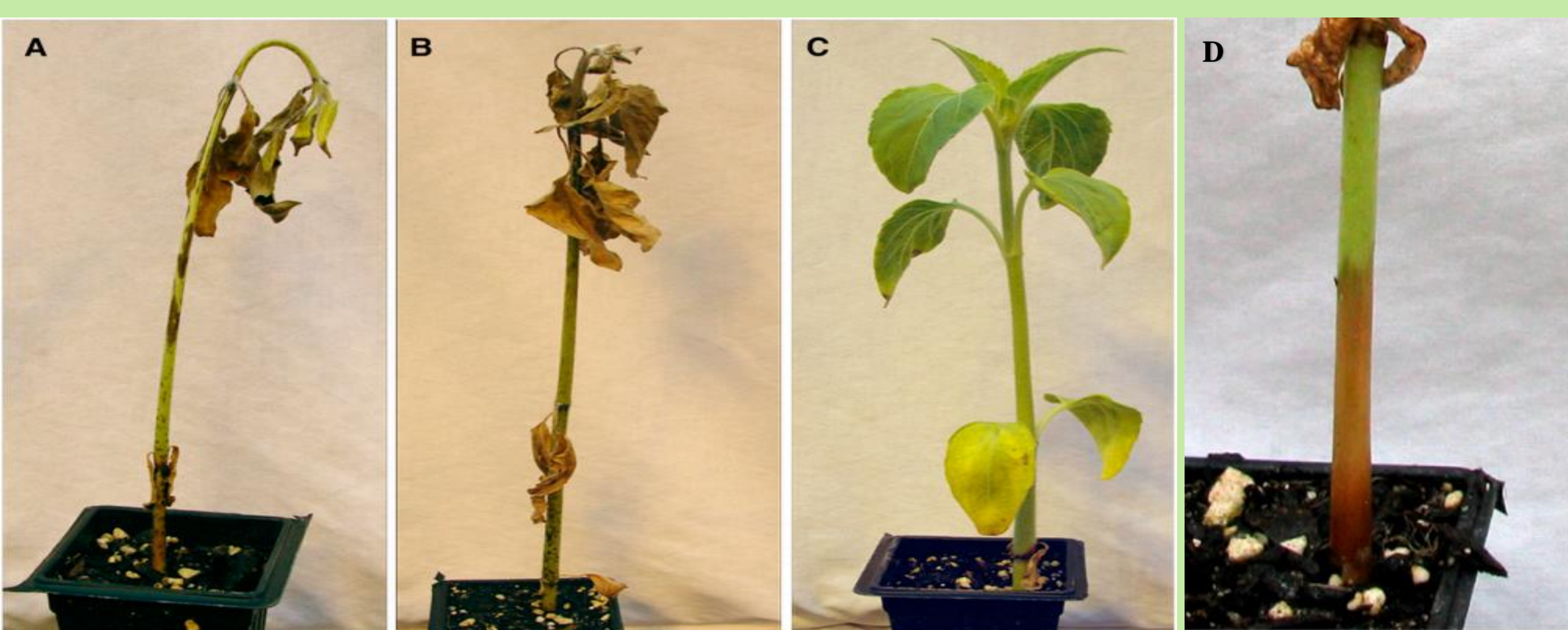


Figure 3: Sunflower BSR symptoms observed in the greenhouse.

A. Plant exhibiting terminal wilt at 14 days post inoculation (dpi). B. Plant exhibiting desiccation at 14 dpi. C. Plant exhibiting few or no symptoms at 14dpi. D. BSR lesion observed after 14 dpi. (2)

Summary and Future work

- Inbred line HA 124 was consistently the most resistant sunflower genotype to all tested *S. sclerotiorum* isolates, suggesting that this line should be prioritized for further characterization.
- Genotype x isolate interactions were significant for sunflower BSR resistance, however, most genotypes identified as resistant in our initial screening were consistently more resistant than the susceptible control.
- Significant variation was observed for *S. sclerotiorum* isolate aggressiveness in causing BSR. Isolates BN166 and BN169 were highly aggressive and may be useful to increase disease pressure in future studies.
- A population of recombinant inbred lines derived from the cross HA 124 x RHA 373 is being developed to facilitate mapping of quantitative trait loci conferring BSR resistance in the highly resistant line HA 124.

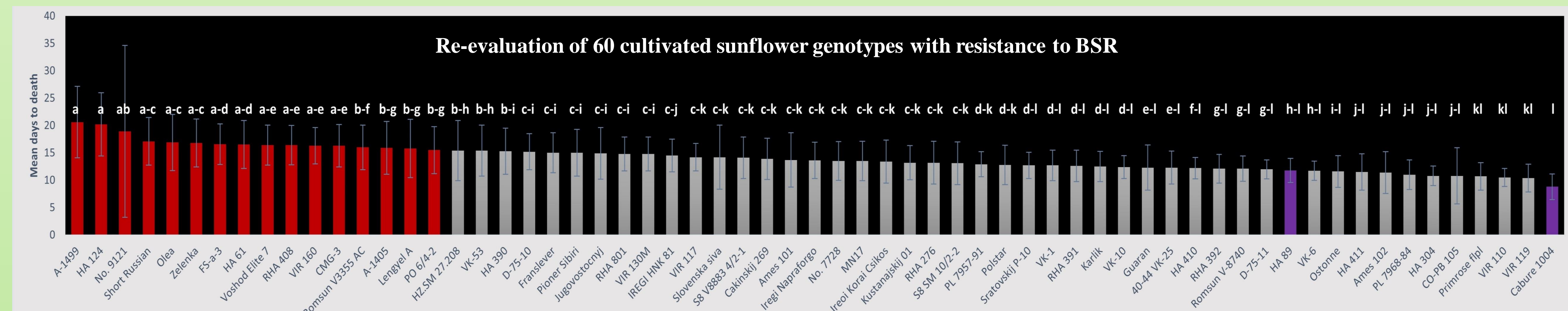
Literature cited

1. Talukder ZI, Hulke BS, Marek LF, Gulya TJ (2014) Sources of resistance to sunflower diseases in a global collection of domesticated USDA plant introductions. *Crop Sci.* 54:694-705.
2. Underwood, W., Misar, C. G., Block, C., Gulya, T. J., Talukder, Z., Hulke, B. S., & Markell, S. G. (2021). A greenhouse method to evaluate sunflower quantitative resistance to basal stalk rot caused by *Sclerotinia sclerotiorum*. *Plant Dis.* 105(2), 464-472.

Results

Re-evaluation of 60 cultivated sunflower genotypes with resistance to BSR

Figure 4: Greenhouse evaluation of BSR resistance for 60 cultivated Sunflower lines. The inbred line check **RHA 801** was included as **moderately resistance** control. Susceptible control includes the **moderately susceptible** inbred line check **HA 89** (purple), **susceptible** open pollinated variety check **Cabure 1004** (purple). Genotypes significantly more resistant than HA 89 and selected for further evaluation are indicated in red color.

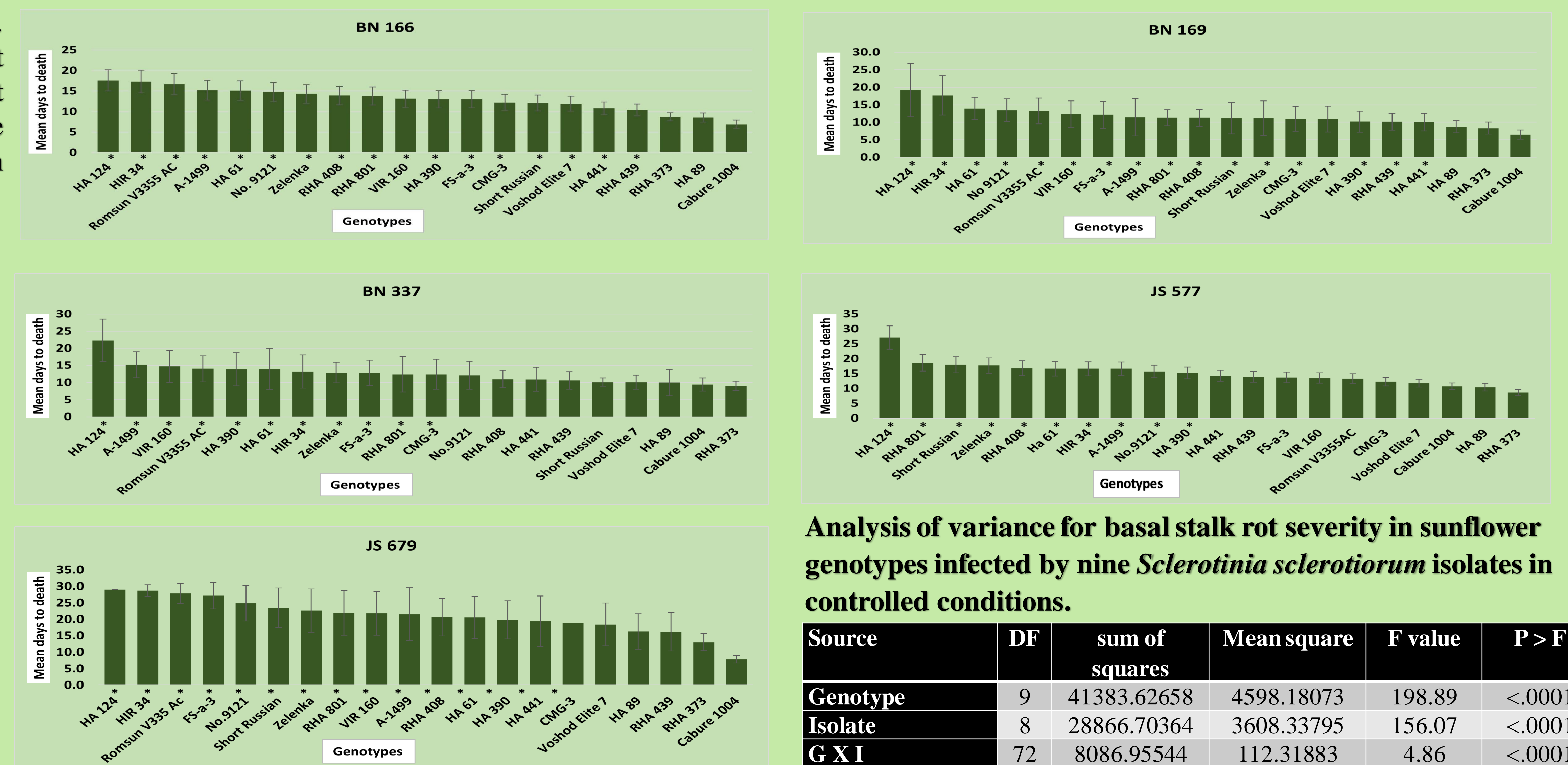


*Lines in statistical groupings indicated by the same letter are not significantly different ($\alpha=0.05$). Error bars represent SD.

Evaluation of selected genotypes with multiple *S. sclerotiorum* isolates to confirm broad-spectrum resistance and assess GXI interactions

- We have previously conducted genotyping-by-sequencing on 227 isolates collected from numerous host plant species throughout the USA to assess genetic diversity among isolates (Belay and Underwood, unpublished).
- Based on this information we have selected five isolates that span the genetic diversity and have been observed to exhibit high or moderate levels of aggressiveness on sunflower stem tissues.

Figure 5: Greenhouse evaluation of BSR resistance for selected sunflower genotypes with diverse *S. sclerotiorum* isolates.



Analysis of variance for basal stalk rot severity in sunflower genotypes infected by nine *Sclerotinia sclerotiorum* isolates in controlled conditions.

Source	DF	sum of squares	Mean square	F value	P > F
Genotype	9	41383.62658	4598.18073	198.89	<.0001
Isolate	8	28866.70364	3608.33795	156.07	<.0001
G X I	72	8086.95544	112.31883	4.86	<.0001

Isolate	DF	Sum of squares	Mean square	F value	P > F
BN 166	9	2797.526227	310.836247	20.66	<.0001
NE 274	9	4407.42315	489.713683	23.02	<.0001
JS 679	9	6708.012038	745.334671	29.72	<.0001
JS 577	9	5384.008472	598.223164	25.34	<.0001
BN 337	9	4878.100625	542.011181	30.88	<.0001
BN 169	9	3247.636408	360.84849	23.71	<.0001
BN 336	9	5246.202379	582.911375	23.41	<.0001
BN 193	9	13800.35454	1533.37273	32.03	<.0001
BN 281	9	3364.354734	373.817193	22.54	<.0001

Genotypes with (*) are significantly more resistant than Cabure 1004. Error bars represent SD.

Acknowledgement - The authors thank the USDA-ARS National Sclerotinia Initiative for research funding.

