SCREENING SUNFLOWER FOR SCLEROTINIA HEAD ROT

Bob Henson, North Dakota State University Carrington Research Extension Center, Box 219, Carrington, North Dakota 58421 USA
E-mail: bhenson@ndsuext.nodak.edu

Tom Gulya, U.S. Department of Agriculture, Agricultural Research Service, Northern Crop Science Laboratory, Fargo, ND 58105 USA
E-mail: gulyat@fargo.ars.usda.gov

Abstract

Sclerotinia head rot (*Sclerotinia sclerotiorum* [Lib.] de Bary) is a devastating disease of sunflower (*Helianthus annuus* L.) and no resistant commercial hybrids are available. A long-term germplasm screening nursery was established in 2000 at the North Dakota State University Carrington Research Extension Center. Entries consisted of production hybrids and experimental lines submitted by private breeding programs. Individual heads were inoculated with ascospores and plots were misted at half-hour intervals to provide favorable conditions for disease development. After several weeks of misting, inoculated heads were evaluated for head rot symptoms. To date, substantial progress has been made in developing the infrastructure (water delivery and misting systems) and methodology (inoculation and evaluation procedures) for conducting a head rot screening nursery. Progress toward resistant commercial hybrids is difficult to assess, since entries vary from year to year and increasingly more entries are experimental lines. However, there are signs of progress. The best of the 82 entries in the first screening nursery in 2000 was used as the resistant check in subsequent years. Every year, several entries, including some confection types, are rated more resistant than the resistant check. Promising germplasm does exist, in both oilseed and confection types. Further testing is needed to confirm these observations and to evaluate new materials. However, variability of disease reaction within plots is still quite high and more research is warranted on the methodology of conducting a head rot screening nursery to optimize labor and land inputs and to increase the precision of the results.

Introduction

Sunflower is highly susceptible to *Sclerotinia sclerotiorum* (white mold) and is unique among broadleaf crops in that infection occurs through the root system (stalk rot) as well as via airborne ascospores. All three types of infection (stalk rot, mid-stalk rot, and head rot) may devastate the crop. However, head rot may result in the most severe production losses, due to contamination of fields and harvested grain by sclerotia in addition to yield losses. With the continued general shift toward more broadleaf (Sclerotinia-susceptible) crop production and continued wet weather in some sunflower areas of the Northern Plains, the threat of Sclerotinia infection is increasing.

No current commercial sunflower hybrids are resistant to Sclerotinia and most are highly susceptible. Public and private institutions and growers have placed a high priority on identifying resistance and head rot screening nurseries are routinely conducted in Argentina and France. These experiences indicate that inoculation with ascospores is more reliable than with mycelia (T. Gulya, personal communication). An inoculum concentration of 5,000 ascospores/ml solution is common, as are 1-row plots. The number of replicates used varies from one to three and row length extends up to 25 feet (7.6 m). Most researchers evaluate disease incidence (percent of plants showing infection), while some also measure severity (degree of damage on infected plants).

Prior to this project, research with sunflower head rot in the Northern Plains was limited to evaluating disease under conditions of natural infection and studying management techniques to minimize disease development. Although severe outbreaks of head rot occurred in both 1999 and 2000, conditions for the development of epidemics normally occur once every four or five years in this region. To assure yearly progress in disease screening, a misting system was designed and constructed to favor disease development. The establishment of the head rot screening nursery in 2000 marked the beginning of a concerted effort to identify genetic resistance in the U.S.A. This work consisted of a three-pronged approach: 1) Evaluate commercial sunflower hybrids and experimental lines for susceptibility to Sclerotinia head rot, 2) Screen USDA-ARS conventional breeding materials, interspecific crosses, and exotic germplasm for resistance, and 3) Develop methodology for conducting an effective and efficient head rot screening nursery.

Materials and Methods

Entries for the head rot screening nursery were solicited from commercial seed companies and compared to susceptible and resistant checks, which were constant every year. One-row plots were 2.5 feet (76 cm) wide x 25 feet (7.6 m) long and were sown in a randomized complete block design with 1, 2, or 3 replicates, at the discretion of the seed company. Participating companies have also had the option of maintaining the results of their entries confidential or making them available to the general public. Performance data for all hybrids were distributed to each company which submitted entries. Hybrids were identified with a trial code and only those entries submitted by the specific company were identified by the true name or number. A seeding rate of 60,000 seeds/acre (148,200 seeds/ha) was used and the population was thinned to 20,000 plants/acre (49,400 plants/ha) after emergence. In 2001, hybrids were grouped by relative maturity (early, medium, and late). A windbreak of border plants was sown on all sides of the experimental area.

At the onset of flowering, when the anthers of the first disk flowers senesced, the faces of individual heads were artificially inoculated with approximately 20,000 ascospores/head dissolved in non-chlorinated water. Ten heads per plot were inoculated when sufficient heads were at the desired stage of flower development. Non-inoculated plants were broken below the head and the first plant in the row was marked with spray paint. End plants in the row were not inoculated. Due to differences in maturity among the entries, at least three inoculation dates were generally needed to inoculate all plots.

Immediately after inoculation, the plot area was misted with well water to favor disease development. The misting system consisted of PVC pipe, black polyethylene hose, plastic tees and adaptors, 6.6-foot (2 m) PVC risers, and mist sprinkler heads. Lines were laid 15 feet

(4.6 m) apart with a similar distance between risers in a line. Risers on adjacent lines were staggered 7.5 feet (2.3 m) to favor optimum coverage. The plot area was divided into zones of equal numbers of risers. The size of the zones was determined by available water pressure. The misting system was controlled by an automatic timer to mist each zone independently for approximately three minutes every half hour, 24 hours / day, until plants were evaluated. The duration of misting cycles was adjusted as weather conditions (relative humidity, temperature) changed.

Approximately five weeks after inoculation, inoculated heads were evaluated for disease symptoms. The following rating scale has evolved over the course of this project: 0=No symptoms; 1= 0–12.5% of head showing symptoms; 2= 12.5–25% of head showing symptoms; 3= 25–50 % of head showing symptoms; 4= 50–100% of head showing symptoms; and 5 = 100% of head showing symptoms.

Disease incidence was calculated as the percent of inoculated heads showing symptoms. A weighted disease severity rating was calculated as follows: Severity = [(# plants rated 1 x 0.0625) + (# plants rated 2 x 0.1875) + (# plants rated 3 x 0.375) + (# plants rated 4 x 0.75) + (# plants rated 5 x 1.0)] / # inoculated plants showing symptoms.

Results

At the Carrington site in 2000, artificial inoculation and misting, coupled with favorable weather conditions, resulted in good disease development (Table 1; Henson, et al., 2001). Disease was rated at four and six weeks after inoculation. The correlation between ratings at the first and second evaluations was 0.995 and very highly significant, suggesting that one evaluation may be sufficient. Results were distributed to cooperators and positive feedback was received regarding the project. However, plant breeders expressed concern over the consistency of data across future years. Also, natural disasters (e.g., hail) could destroy the plots and a second site with misting was suggested at South Dakota State University, Brookings, South Dakota.

At the onset of the 2001 growing season, the well and pump used for the misting system were renovated to improve efficiency and dependability and to facilitate periodic monitoring of performance (Hla, 2001; Henson, 2001). The underground water delivery system was expanded to provide more access points and permit at least a 5-year field rotation. A long crop rotation is essential to the long-term success of this project, since a tremendous amount of disease inoculum (sclerotia) is left in the field and consequent stalk rot could significantly compromise the results of future head rot evaluations.

In 2001, commercial hybrids were again screened in Carrington (Henson et al., 2002) and a subset of these entries (at the discretion of the seed companies) was also evaluated in Brookings (Draper and Ruden, 2002). August 2001 was quite dry. Benefits of the misting system were realized when non-misted thesis plots in Fargo produced no useable data, while those at Carrington did.

The 2002 and 2003 growing seasons in the Carrington area were again relatively dry, with low levels of natural infection in production fields. Although the 2002 data was more variable, satisfactory data was recorded in both years.

Table 1. Disease incidence (% of inoculated plants showing infection) in the sunflower head rot screening nursery, North Dakota State University Carrington Research Extension Center.

	2000	2001 Early-maturing	2001 Medium-maturing	2001 Late-maturing	2002	2003
Entries	82	11	38	35	55	33
Mean	57	40	35	33	61	28
Range	17-100	10-77	0-80	0-100	17-100	0-73
C.V. (%)	40	47	59	63	51	46
P-value	< 0.001	0.012	0.008	0.004	0.835	< 0.001

Discussion

The anticipated outcome of the head rot screening project is a long-term nursery, which will provide a reliable evaluation of sunflower hybrids and germplasm for Sclerotinia susceptibility. This information is of value to growers in selecting hybrids to plant. The results are of benefit to plant breeders in identifying parental material and promising experimental lines and should facilitate the development and release of hybrids with increased tolerance/resistance to Sclerotinia.

Progress toward resistant commercial hybrids is difficult to assess from the results of the screening nursery, since entries vary from year to year. Also, more and more entries are experimental lines and not released hybrids. A line may show promise as a source of disease resistance in a breeding program, but may lack other traits needed in a commercial hybrid. However, there are signs of progress. The best of the 82 entries in the first screening nursery in 2000 was used as the resistant check in subsequent years. Several entries, including some confection types, have been rated more resistant than the resistant check. Promising germplasm does exist, in both oilseed and confection types.

Lessons Learned. 1). The misting system must be extended beyond the plot area to allow for drift on windy days. 2). A weather-resistant system of identifying inoculated plants is essential. Early efforts with paper tags and some colors of spray paint were not durable under the misting system. 3). The misting cycle (minutes on per cycle) must be balanced between maintaining a humid environment and avoiding lodging. The plots are misted, not irrigated. 4). A windbreak is important to reduce drying and maintain a humid environment within the plot area. In 2000, a tall sunflower hybrid provided a functional windbreak until it lodged severely. The current procedure is to plant a 2-row border of sunflower plants surrounded by a 2-row border of tall, silage corn. The entire plot area is surrounded by a 2-row border of sudangrass, which provides a thick and pliable barrier to wind and deer. 5). In 2001, germplasm was grouped by relative maturity. However, no advantages were found in this procedure and the arrangement precluded one statistical comparison of all entries. 6). The correlation between disease rating and disease incidence was highly significant (0.924). However, exceptions do exist and both criteria should continue to be evaluated. 7). The correlation of disease rankings between two and three replicates of data is highly significant (P = 0.947), suggesting that two replicates are sufficient. Although the correlation with one replicate is also high, the risk of losing a plot to a poor stand or predation is too high. 8).

Disease ratings are typically lower with later inoculation dates. This may be an indication of higher resistance in germplasm with longer maturity or may be an artifact of less favorable conditions for disease development later in the season.

Refinements Needed. Additional research is needed on the most efficient and effective methodology for head rot screening. Improved efficiency will permit the evaluation of more germplasm, while maintaining realistic commitments of personnel and infrastructure. Increasing effectiveness will improve the accuracy and precision of the data generated, confidence in the system, and the identification of superior germplasm. The following are refinements needed: 1). Evaluate the benefit of adding a food (sugar) source to the solution of ascospore inoculum. Inconsistent disease ratings are often observed among individual plants within plots. Although this observation may be an artifact of working with a cross-pollinated crop and some experimental lines, it may be due to inoculation failure. Improving the survival of the disease inoculum may improve this inconsistency and should improve overall infection. 2). Reduce the variability among plants and replicates in the nursery. 3). Develop methods of data analysis to maximize the information provided. 4). In collaboration with breeders and user groups, optimize reporting of the results, and 5). Define the appropriate number, frequency and distribution of susceptible and resistant checks.

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