IMPACT OF THE SOYBEAN CYST NEMATODE ON SEEDLING DISEASES OF SUGARBEET

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Soybean cyst nematode (SCN; *Heterodera glycines*) was first reported in North Dakota in 2003 (1) and was also recently discovered in the Red River Valley (RRV) of Minnesota. SCN is now well-established in Cass and Richland Counties of North Dakota and Wilkin, Clay, Norman and Red Lake counties in Minnesota, and is likely to continue to progress northward as long as soybean production continues. The nematode is easily moved from field to field in soil on farm equipment and in harvesting operations. The nematode survives from year to year as eggs within cysts. Although SCN is the most important pathogen of soybean in the United States (3), it is not reported to reproduce on sugarbeets. However, SCN is very closely related to the sugarbeet cyst nematode (SBCN). Indeed, the two species can hybridize to form fertile offspring (2).

When SCN eggs hatch, second stage juveniles (J2) emerge and maneuver through the soil to find a host root (3). Within hours of arriving at the surface of a susceptible soybean root, the nematode has penetrated through epidermal cells and migrated intracellularly to the vascular cylinder of the root. The female juvenile then initiates a permanent feeding site called a syncytium from which it draws nutrients from the plant root for its growth and development. However, when the juvenile arrives at the root surface of a non-host plant, penetration of the root can occur but the syncytium is not established. These larvae will either die or may exit the non-host root.

SCN can build to high populations in field soil in the RRV. In Richland Country, fields have been measured that had average egg densities of over 10,000 eggs/100 cm^3 soil, and spots within fields had levels of 30,000 eggs/100 cm^3 soil. When sugarbeet is planted into SCN-infested soil, the nematode may attempt to penetrate and establish itself in the sugarbeet roots. Such penetration attempts are likely to create lesions on the root surface. In addition, wounded roots may have an altered production of root exudates that attract sugarbeet pathogens. Since wounding of the sugarbeet root is known to increase disease severity for several sugarbeet diseases, the lesions made from entry by SCN might offer convenient entry points for several sugarbeet pathogens. If penetration by SCN were to occur at high levels, there is a possibility it could increase susceptibility of sugarbeet roots to root pathogens such as *Rhizoctonia solani*, *Aphanomyces cochlioides*, *Verticillium dahliae*, or *Fusarium* species, especially in the seedling stage when plants are more susceptible to pathogens. In soybean, SCN is well-known to increase root diseases caused by fungal pathogens.

OBJECTIVES

The objectives for 2010 were to:

1. Determine if juveniles can penetrate sugarbeet root tissue under field conditions
2. Determine if sugarbeet age influences nematode penetration
3. Determine if SCN causes increased susceptibility to soil-borne pathogens

MATERIALS AND METHODS

Females of *Heterodera glycines* (HG type 0) were maintained in the greenhouse on susceptible soybean cultivar Barnes. Mature 30-day-old cysts were crushed to remove eggs for inoculation.

A randomized complete block design was constructed in a microplot under field conditions. Three treatments levels of SCN (10,000 eggs/100cm^3, 5,000 eggs/100cm^3 and control) were split among three cultivars (ACH 17, SVDH M832224 and SVDH 46519) in a total of 5 replications. Soil determined to be free of SCN was collected, treated with SCN and placed into 12 liter pots and placed into the microplot. Harvesting of the sugarbeets took place over three time points at periods of approximately one, two and four months post planting. Roots were harvested and cleaned to remove excess soil, then they were freeze dried, ground to a fine powder and genomic

DNA was extracted. Polymerase chain reaction (PCR) was performed on plants from all three timepoints with SCN-specific primers.

To determine at what age sugarbeets are most susceptible to SCN penetration, cultivars M832224 and 0957-22 (SESVanderHave) were planted every week in the greenhouse. After 7 weeks, approximately 10,000 SCN eggs were added to the soil of each pot. The plants were then transferred to a growth chamber where the temperature was held constant at 27ºC. The plants were incubated in the growth chambers for 10 days. After the incubation period, the roots were removed from the soil, cleaned and freeze dried. DNA was extracted using a DNeasy kit (Qiagen, USA). Quantitative PCR (qPCR) was then performed with primers designed specifically for SCN and sugarbeet actin. The experiment was repeated twice.

To determine if pre-infection by SCN increases disease severity from *R. solani* AG 2-2 IIIB, plants of the cultivar 0957-22 were started in the greenhouse. At three weeks of age, half of the total plants in the experiment were inoculated with 10,000 SCN eggs. Twenty-four hours later, two barley kernels containing inoculum of *R. solani* AG 2-2 IIIB were placed 2 cm from the root on the surface of the soil. The plants were then incubated for 10 days in growth chambers where the temperature was kept constant at 27ºC and humidity was kept at 50%. After 10 days the plants were pulled from the soil and were rated for *Rhizoctonia* disease using a 0-7 rating scale. The experiment was repeated three times.

RESULTS

SCN penetration under field conditions

It was determined through PCR that SCN could be found in all three time points. The bands shown in Figure 1 signify the presence of SCN found within the roots; however, intensity of the band brightness may not correlate to amount of SCN present.

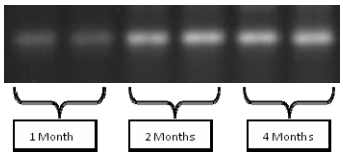

Figure 1. PCR confirmation of SCN in sugarbeet roots at selected timepoints.

Influence of sugarbeet age on SCN penetration

Plants from the variety of 0957-22 showed the greatest amount of infection when inoculated at 1 and 2 weeks of age (Fig. 2). Since plants were anywhere from 2 to 4 weeks of age at the time of tissue harvest, the most susceptible time points for SCN penetration of 0957-22 are between one and four weeks of age. Cultivar M832224 had a slight increase in SCN penetration at 4 weeks of age (Fig. 2).

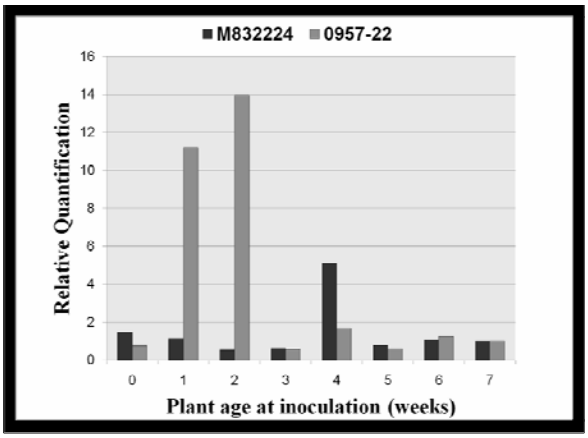

Figure 2. Influence of age on SCN penetration.

Influence of SCN on *R. solani* disease severity
Plants pre-treated with SCN and then *R. solani* had almost twice the necrosis than plants treated with *R. solani* alone (Fig. 3). However, there was significant variability in disease ratings from both treatments.

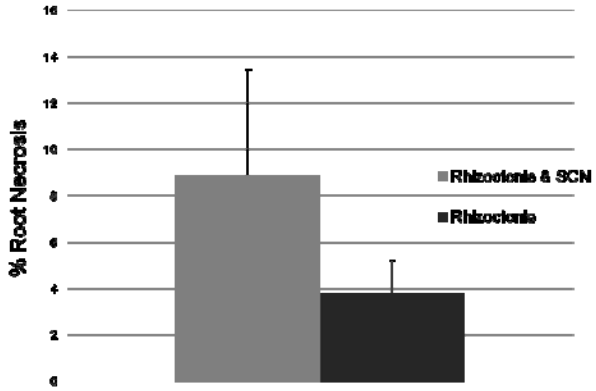

Figure 3. Influence of pre-infection by SCN on *Rhizoctonia solani* disease severity.

DISCUSSION

In this study, we found SCN in sugarbeet tissue grown under field conditions under all three selected time points (Fig. 1). The finding that SCN can be found in sugarbeet tissue four months after planting suggests that SCN is likely to be found in root tissue under field conditions in grower fields.

We were also interested to see which growth stage is the most susceptible to SCN infection. To determine this, we stagger-planted plants every week for 7 weeks. At week 7, all plants were inoculated with SCN and SCN quantification was determined 10 days later. Interestingly, the cultivar M832224 that is sugarbeet cyst nematode- (SBCN-) susceptible did not accumulate as much SCN as the sugarbeet tolerant cultivar 0957-22 (Fig. 2). This suggests that tolerance to nematodes in sugarbeet cultivars slows down nematode reproduction, but does not impart penetration resistance. Alternatively, the resistance associated with 0957-22 may be specific to SBCN.

We found that pre-infection by SCN imparts an increase in disease susceptibility to *R. solani*. In every repeat of the experiment, there was an obvious increase in disease in SCN-pre-treated plants. However, variability in our results is a concern. Future work will focus on adjusting the time points of inoculation and harvest, which we feel will decrease variability. Since SCN appears to increase susceptibility to *R. solani*, we are also interested if this is the case with Fusarium and other soil pathogens of sugarbeet. We have been using 10,000 SCN eggs/100 cm^3 soil as our benchmark level to assess if SCN increases susceptibility. However, fields with 30,000 SCN eggs/100 cm^3 have been found in Richland County. Therefore, we would like to determine the threshold level of SCN eggs that imparts a significant increase in susceptibility to *R. solani* and other sugarbeet pathogens. Finally, we are interested if pre-infection by other root "nibblers" such as springtails and sugarbeet root maggots also have a similar effect on sugarbeet diseases.

LITERATURE CITED

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