

BEYOND PATHOGENS:

Expanding Metagenomic Insights to Soybean Cyst Nematode





Soybean Cyst Nematode (SCN) is a devastating pest for soybean growers, though it is possible to detect and manage. The most common test for SCN is performing egg or cyst counts from soil samples. Advances in DNA sequencing technology have made shotgun metagenomics a valid alternative that can test for SCN concurrently with hundreds of other pathogens as well as provide functional nutrient data.

Soybean Cyst Nematode (SCN) is considered the most significant soybean pest in the United States – causing annual yield losses of over \$1 billion.¹ The nematode, *Heterodera glycines*, is difficult to detect before serious damage occurs and can be difficult to manage due to its long persistence in the soil and broad host range. If infestations go undetected, yield can drop slowly for years as the SCN population increases.

The current industry standard for diagnosis of SCN infestation is to perform egg or cyst counts from a soil sample. This method frequently yields false negative results due to human error as eggs are overlooked or misidentified during visual inspection, and turnaround time is extensive. Because there are no consistent visual symptoms on the crops, diagnostics for SCN cannot be scaled up using remote sensing scouting technologies.

Improving Pest Diagnostics with DNA Technology

A scalable way to improve the detection of SCN is to use molecular or DNA-based diagnostic techniques. In human health, a PCR test is used to diagnose COVID-19. Though this method is sensitive and accurate, it is limited to only a few specific targets at a time. The COVID-19 PCR test can be performed concurrently with influenza A and B; however, doctors would need to order additional tests for other respiratory illnesses or diseases. Similar to these PCR tests, qPCR (quantitative PCR) is commonly used to diagnose plant pathogens, including SCN.^{2,3} If growers test for SCN using qPCR (or egg counts), they still need to order separate tests for data on other pests, as well as beneficial microbes, fertility data, and soil chemistry.

Shotgun metagenomics (also known simply as “metagenomics”) is a non-targeted method for sequencing all DNA in a sample, which provides a comprehensive analysis of soil biology. Traditionally, metagenomics has only been used for analysis of microorganisms such as fungi and bacteria. However, DNA from larger organisms like nematodes and insects can be detected with this technique as well.

When used to detect SCN, metagenomics measures DNA that originates not only from eggs, but other sources as well, such as juveniles and adults. Additionally, it is sensitive enough to detect SCN in soils in cases where egg counts are low or undetectable. When examining samples that have both metagenomic and egg count data, we have found that metagenomics can reliably determine the presence or absence of SCN in soil samples (Figure 1). Additionally, the turnaround time for metagenomic analysis is significantly faster than egg counts, with results delivered in days rather than months.

Trace Genomics offers metagenomic analysis alongside soil chemistry through the TraceCOMPLETE package. This product not only provides actionable data for managing SCN, it also includes quantitative data for more than 225 additional pests, biological nutrient cycling indicators, and beneficial symbiotic microbes. Furthermore, this abundance of data is packaged into reports that include interpretation and guidance that can be used to make management and product placement decisions.

REFERENCES

1. Wrather, Allen, and Melissa Mitchum. “Soybean Cyst Nematode: Diagnosis and Management.” University of Missouri Extension, Aug. 2010, extension.missouri.edu/publications/g4450.
2. Hart, Patrick. “Application of Real Time PCR for Detection and Identification of Soybean Pests in Michigan.” Michigan State University Archives, 2004, www.canr.msu.edu/field_crops/uploads/archive/ID%20soybean%20pests%20GR05-004.pdf. Accessed 15 Aug. 2023.

It should be noted that for all the methods mentioned above (egg counts, qPCR, and metagenomics), false negative results are possible if SCN is present in a field but not collected in the tested soil samples. Since SCN distribution across a field is not uniform, several samples should be collected across a field with multiple cores per sample taken from within and between rows. More detailed sampling guidance documentation is provided by Trace Genomics to reduce the chance of false negatives.

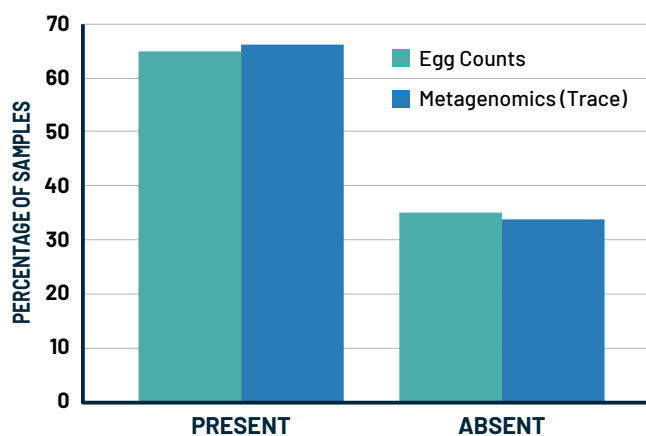
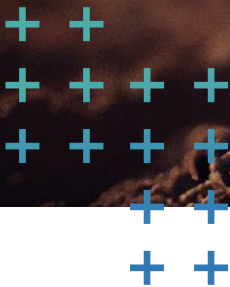


Figure 1. Comparison of SCN detection results using traditional egg counts and metagenomics. Metagenomics was performed by Trace Genomics, and egg counts were done on the same soils at Iowa State University. Bars show whether a method determined that SCN was present or absent (teal: egg counts, dark blue: metagenomics).

Interpreting SCN Results from Metagenomics

While metagenomics has been validated for the detection of SCN from soil samples, the quantity cannot be directly translated into egg counts because metagenomics detects DNA from other sources in addition to eggs. Quantities of SCN DNA from metagenomics can still be used as a measurement of SCN density in the soil. These values can indicate whether a field is infested with SCN, so the appropriate management actions can be taken. Additionally, the measurement from metagenomics is also a valid way to evaluate whether implemented management strategies were effective by tracking whether the quantity of SCN increases or decreases over time.

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EXPERIENCED AGRONOMISTS
RECOMMEND TAKING ACTION
IF ANY LEVEL OF SCN IS
FOUND IN YOUR FIELD.**



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