

Objective 1: Establish baseline knowledge of microbial communities associated with SCN cysts in NY state

The Fall 2023 SCN survey sampled soil samples from 33 fields across 10 counties in NY (Table 1) identified 7 fields in three counties (Tioga, Montgomery, and Herkimer), with only three fields, two in Tioga County and one in Montgomery County, showing substantial egg counts above 1000. These results suggest SCN is still in the early stages of spread in NY but has spread and reached more northern counties (Montgomery, Herkimer) in the middle of the state. From DNA metabarcoding of ITS for fungal and 16S for bacterial communities, we identified a set of core bacterial and fungal taxa found across a majority of soil samples in NY state. We also found differences in the microbial communities between soil in SCN infested versus uninfested fields, suggesting that the presence of SCN may be causing a change in microbial communities. In the presence of the SCN pathogen, soil communities have been shown to accumulate nematode parasitic microbes over time.

To sample these microbes, the last (July 31) progress report described the results of culturing ~200 fungi and ~200 bacterial cultures from the SCN infested soils. This past quarter, we have focused on sequencing these microbes to classify them to genus and identify those that may be nematode parasites for further testing in Objective 2. We have completed sequencing and identification of 100 fungi to genus, prepared remaining fungi for sequencing, and are in the process of amplifying the 16S for bacterial sequencing. For fungi, we found that the largest number of fungal isolates belonged to the fungal order Hypocreales, which is known to harbor many nematode parasitic fungi (Fig. 1). Other orders identified in our culture collection known to have nematode parasites include Mortierellales, Eurotiales, and Pleosporales.

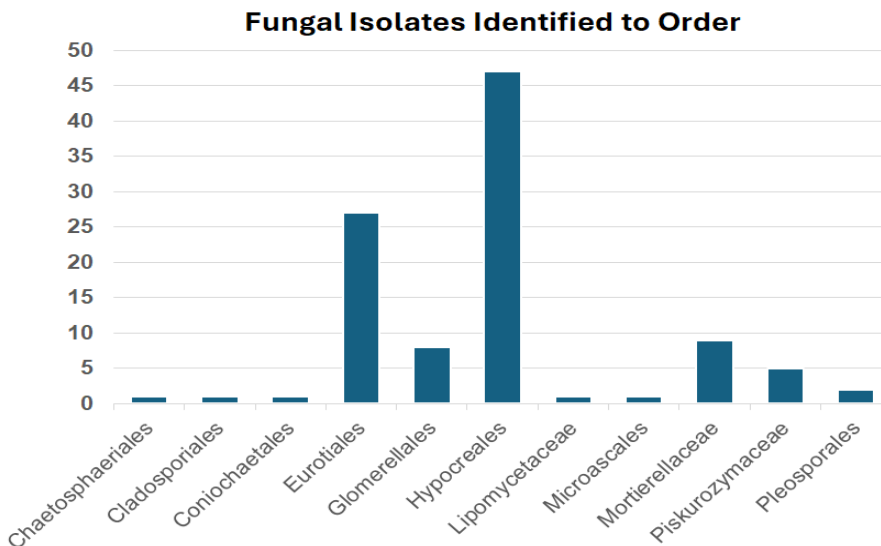


Fig. 1. Fungal isolates identified to order level

Among the Hypocreales, we identified fungal isolates belonging to twelve different genera. Many of these genera (*e.g. Fusarium, Pochonia, and Purpureocillium*) have been documented in the literature and in our previous research to contain nematode parasites and/or produce compounds toxic to nematodes and are now our top candidates for testing in Objective 2. While other genera are not as well studied, they could have activity and a representative of these genera will be screened for nematode antagonistic properties.

Objective 2: Screen fungi for antagonism of the soybean cyst nematode

1.2 Screen fungi for production of nematicidal and hatch inhibitory metabolites

We have screened filtrates from fungi isolated from soil using J2 toxicity assays. Out of 50 screened, 5 have shown significantly greater mortality of J2 exposed to the raw fungal filtrates of a fermentation broth compared to the media (1/4 Czapek-Dox broth) control. In the previous reporting period, one of these isolates whose raw filtrate showed high toxicity to J2 in bioassays was grown up in rice media for one month. Raw extractions using a 1) water (RW), 2) a relatively polar solvent butanol (RB), and two nonpolar solvents, ethyl acetate (RE) and hexane (RH), as well as a distilled water control, were tested in toxicity bioassays with J2 worms. Results showed that the ethyl acetate fraction (RE) had significantly higher mortality than the water extract. This period, the raw extracts have been further chemically fractionated to isolate pure compounds and these fractions are currently being retested in bioassays. Results thus far show that the butanol extracts have the highest hatch inhibition. We have also grown up the additional four isolates whose fungi whose filtrates showed activity in rice media under the same conditions and prepared raw extracts that are currently being tested.

Table 1. Soil sampling for SCN survey (Fall 2023) showing 33 fields sampled in 10 counties across NY.

Sample number	County	# Cysts	Viable Eggs Counts
42H	Chautauqua		
42L	Chautauqua		
43	Chautauqua		
44	Chautauqua		
7	Chautauqua		
15	Cortland		
16	Cortland		
17	Cortland		
18	Cortland		
19	Cortland		
10	Cortland		
11	Cortland		
45	Erie		
46	Erie		

38	Herkimer		
D	Herkimer	2	44
33	Madison		
28	Montgomery	19	2080
30	Montgomery		
40	Montgomery		
41	Montgomery		
B	Montgomery	5	210
A	Montgomery		
35	Otsego		
37	Otsego		
C	Saratoga		
47	Tioga	20	1400
48	Tioga	4	91
49	Tioga	5	51
50	Tioga	87	2100
12	Tompkins		
14	Tompkins		
20	Tompkins		