

## **Geddes lab min-term progress report to North Dakota Soybean Council: Potential for combatting iron deficiency chlorosis with the soybean microbiome**

### **a. Objectives of the Research**

Objective 1) Evaluate the restructuring of the soybean microbiome composition during iron deficiency chlorosis.

Objective 2) Measure the potential for iron release by the soybean microbiome using a colorimetric chrome azural S assay.

Objective 3) Optimization of a greenhouse assay for measuring reduction in IDC by microbial inoculants in soybeans.

### **b. Completed Work**

In collaboration with Dr. Carrie Miranda, we have collected root samples and scored IDC levels of soybeans grown in four locations in ND with varying iron deficiency chlorosis condition to be used for analysis of microbiome composition during IDC. These included Leonard (IDC +++), Colfax (IDC ++), Casselton (IDC +) and Prosper (IDC -) (Figure 1). A total of 64 samples were collected with three soybean plants each, representing two resistant varieties (A11 and Rolette) and two IDC sensitive varieties (ND17009 and ND16). The rhizosphere (zone of soil around the root) of soybeans from each location was isolated and separated from the endosphere (inside the root) fraction for processing for microbiome analysis (Objective 1). We also stored glycerol stocks from the rhizosphere microbiome to be used in the chrome azural S assay (Objective 2).

We have successfully optimized an in house protocol for sequencing of the bacterial 16S gene by next-generation sequencing using the Illumina MiSeq in the department of Microbiological Sciences. The DNA from IDC rhizosphere samples has been isolated and is ready to be sequenced to determine the bacterial community structure using the optimized protocol.

We have coordinated with Dr. Jay Goos to adapt his greenhouse IDC assay towards measuring the microbiome effect on IDC in the greenhouse. A pilot study that includes 160 soybean plants is currently ongoing in the greenhouse to establish the optimal conditions for measuring reduction of IDC by microbes from the soybean microbiome (Objective 3).



Figure 1. IDC trial sampled at Leondard

### c. Preliminary Results

The observed IDC severity from the four varieties in the four selected locations showed the expected IDC response, indicating we have collected a fantastic dataset for the upcoming microbiome analysis (Figure 2).

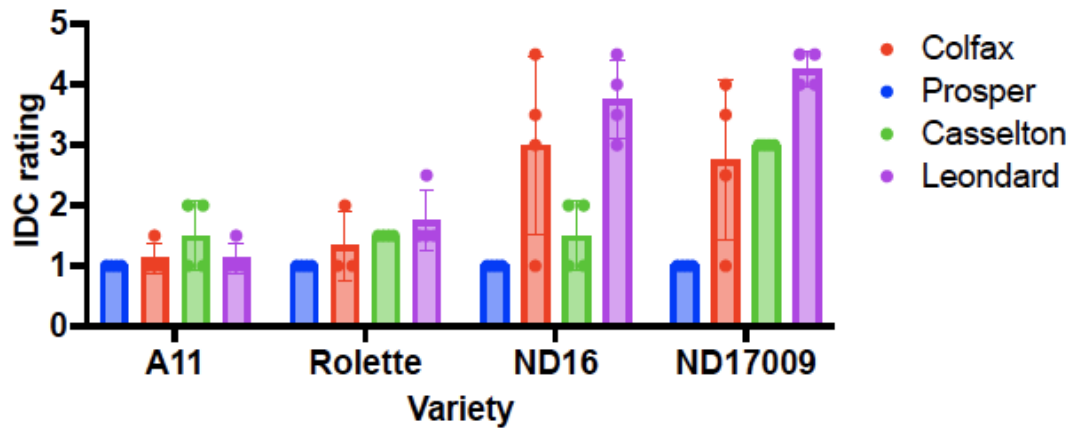


Figure 2. IDC severity rating from plants harvested for microbiome analysis.

### **c. Work to be Completed**

All three objectives are yet to be finalized:

Objective 1: To complete the microbial community profile from field-grown soybeans we will extract DNA from the soybean endosphere samples to combine with the previously extracted rhizosphere samples. These samples will be sequenced for bacterial microbial community structure using our optimized protocol. Afterwards data analysis will be performed to identify microbial taxa that may be recruited by the plant during IDC.

Objective 2: Samples have been collected but the chrome azural S assay still needs to be performed using preserved microbes from the soybean rhizosphere.

Objective 3: Once the IDC greenhouse assay has been optimized, it will be used to assess the potential of microbes from the soybean microbiome to reduce IDC.