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MPSG ANNUAL EXTENSION REPORT

PROJECT TITLE: Greenhouse screening of soybean varieties for resistance to iron deficiency chlorosis

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PART 1: PRINCIPAL RESEARCHER

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PART 2: EXECUTIVE SUMMARY

Evaluation of soybean breeding/genetic populations for iron (Fe) deficiency chlorosis (IDC) resistance is mainly based on foliar chlorosis symptoms. The ability to differentiate IDC susceptible and resistant plants is therefore critical for the success of an IDC breeding program. Expression of IDC is affected by many soil and environmental factors. Environmental factors that can confound IDC phenotyping include heterogeneity in soil composition, rainfall patterns, and temperature etc. These factors cause spatial and temporal variations in Fe availability so that it can change dramatically even within one field and one growing season. Moreover, even the visual chlorosis symptoms can show interactions with environment. In 1992, Chaney et al. outlined a detailed procedure for chlorosis screening using nutrient solutions which can be used routinely by breeders. Some advantages associated with using nutrient solutions for IDC screening are, (i) a high severity of chlorosis can be induced, (ii) confounding effects of environment such as soil heterogeneity can be avoided, (iii) screening can be done year around, and (iv) genotypic evaluations take a short time for each cycle (~1 month). The hydroponic protocol has been successfully set up at Morden using 21 soybean materials. Resistant (R) and susceptible (S) lines are differentiated at various growth stages. The protocol and selected R and S lines as checks will be used for large scale screening of soybean breeding materials in 2016. Resistant materials will be used for crossing for early-maturing variety development.

PART 3: PROJECT ACTIVITIES AND PRELIMINARY RESULTS

Methodology: A protocol published by Chaney et al (1992) was used to establish IDC screening setup at Morden-AAFC. Table 1 shows the list of 21 soybean varieties used for IDC screen in the current update. Figure 1 shows the procedure used for IDC screening. Soybean seeds were germinated in wet paper towels to obtain seedlings. Four one-week-old seedlings per genotype were transferred to buckets containing Diethylene triamine pentaacetic (DTPA) chelated nutrient solution. Holes in plexi-glass bucket covers were used to suspend the seedling roots in solution and support the stems for upright growth. The hydroponic nutrient solution contained all essential macro- and micro-nutrients required for proper plant growth except Fe. To create Fe deficiency, 25µM Fe-DTPA and 25mM $NaHCO_3$ was also added to each bucket. Sodium bicarbonate can cause excessive alkalinity in the nutrient solution. Therefore, to maintain pH for proper plant growth, a 3% CO_2 mixture was bubbled into the buckets using a manifold assembly. The bubbling of gas mixture also helped in circulating nutrients in the buckets. Nutrient-solution-volume loss due to evaporation was compensated by adding distilled water throughout the growth period. A visual rating scale was used to score IDC symptoms. Rating Scale was: 1=full green; 2=mild chlorosis; 3=full yellow leaves but veins remain green; 4= yellow with little green in the vein; 5=full yellow with necrosis. Resistant (Clark) and susceptible (Iso-Clark) checks were introduced from the Iowa State University and used in the tests. Early maturity soybean cultivars (Jim, Glacier, Maple Ridge and Maple Presto) were also included as checks.

Progress: The above shown setup (Figure 1) was successfully used to screen early maturity soybean accessions and varieties in Fall 2015. Table 1 shows the results of the screen. The screen was successful in differentiating IDC resistant cultivars (low scores) from sensitive cultivars (high scores). Ratings were recorded at various early stages of growth. The screen, starting from seeds to the last rating was completed in only 31 days.

The IDC symptoms occurred as early as the first trifoliate stage, ranging from 1 (no chlorosis) to 3 (highly yellowish plants). The susceptible cultivars such as Glacier and Iso-Clark showed high rating scores consistently during the screening, and can be used as susceptible checks in the future tests. Meanwhile, Maple Ridge and PI449456 B were consistenly resistant from day 21 to day 31. The IDC symptoms of some cultivars recovered as plants grew, but most cultivars showed progressively development of IDC as plants grew bigger.

Experiments with various growth mix, sand, and soils collected from Emerson soybean field were also conducted using various size of pots and watering with nutritional solutions deficient of Fe. Plants did not develop any significant IDC symptoms before flowering.

Future plans: In 2016, we will continue to use greenhouse IDC screening procedure to screen both popular and upcoming varieties in Manitoba. This screen will help in identifying cultivars which could be highly sensitive to IDC under the favourable field conditions. The screen will also help in choosing parents for making crosses in order to develop future varieties.

Reference:

Chaney, R. L., B. A. Coulombe, P. F. Bell, J. S. Angle. (1992). Detailed Method to Screen Dicot Cultivars for Resistance to Fe-Chlorosis Using FeDTPA and Bicarbonate in Nutrient Solutions. J. Plant Nutr. 15(10): 2063-2083.

APPENDIX

Table 1: Chlorosis scores of early soybean accessions in hydroponics IDC experiment. Hydroponics nutrient solution with 25µM FeDTPA and 25mM NaHCO3 was used to create Iron deficiency stress.

Sr. No.	Early Soybean Varieties	Accession No.	Chlorosis score rating					
			Day 21	Day 24	Day 25	Day 27	Day 31	Average Score
			1st trifoliate	2nd trifoliate	2nd trifoliate	2nd trifoliate	2nd trifoliate	
1	Glacier (Check)		3	3.5	3.75	3.75	3	3.4
2	Jim (Check)		2.25	1.5	1.25	1	1	1.4
3	Maple Presto		2.5	1.75	1.5	1	1	1.55
4	Maple Ridge (Check)		1	1.5	1.25	1	1	**1.15**
5	Kievskaja 91	PI 567216 A	1	2	2.5	2.5	2	**2**
6	Tercinskaja 24	PI 507704 A	2.75	2	2.5	1.75	1.8	2.16
7	Maple Amber	PI 548592	2.75	2	2.25	2	1.75	2.15
8	Jiu feng No. 4	PI 549080	2.75	3.5	3.5	3	2.5	3.05
9	Jug 30	PI 567218	1.5	2.5	2.5	3	3	2.5
10	McCall	PI 548582	2.25	2.5	2.5	3	3	2.65
11	Kokuso	PI 227327	1.75	2	2.25	2.75	3	2.35
12	VNIIS 2	PI 507711 A	1.75	1.75	1.5	2.5	3	2.1
13	Sansinduscaja	PI 437920 A	1.75	2.75	2.75	2.5	2	2.35
14	Daksoy	PI 602896	1.5	1.5	1.5	1.5	2	**1.6**
15	(Yantarnaya)	PI 561282 B	2.75	3	2.5	3	2.5	2.75
16	(Bei 77-6177)	PI 449456 B	1	1.5	1.25	1.5	1.5	**1.35**
17	Pagodo	PI 548398	2.5	3.5	4	4	4	3.6
18	S150	PI 548648	1.5	3.25	3	3.75	3.5	3
19	S160	OAC Prudence	2.25	1.75	1.5	2	2	**1.9**
20	Clark (IDC Check)		1.5	2.5	3	3.25	3	2.65
21	Iso-clark (IDC Check)		3	3.5	3.75	4	3.75	3.6
Rating Scale: 1=full green; 2=mild chlorosis; 3=full yellow leaves but veins remain green; 4= yellow with little green in the vein; 5=full yellow with necrosis								

Figure 1: Depiction of the greenhouse IDC screening procedure. A. Seeds are germinated in wet paper towels to produce seedlings for transplantation; B. One-week-old seedlings are transplanted into the buckets containing nutrient solution; C. Nutrient solution is aeriated with 3% CO_2 to circulate nutrient solution and maintain pH; D. Gas flowmeter is used to generate 3% CO_2 for aeriation; E and F. IDC sensitive cultivar showing IDC symptoms under iron stress (E) 25 µm FeDTPA and healthy growth under non-stress (F) 100 µm FeDTPA.