- Agenda: Integrating next-generation technologies for blackleg and soft rot management in potato. Progress and reports. 2019
- Thursday November 7
- Ballroom IIIC, Intercontinental Hotel, Minneapolis airport
- Walkway entrance by Gate C26
- 10:00. Welcome, housekeeping announcements, introductions
- 10:15 to 12:00. Short five to ten minute reports by project scientists, graduate students, post docs on current projects and preliminary results Discussion and questions welcome and encouraged after each presentation
- Presentations grouped by subject for focused discussions
    - Genome/Genetics
    - Epidemiology/Detection
    - Host Resistance/Genetics
- 12:00 to 1:30. Farm House buffet lunch in the meeting room, informal discussions, check emails, etc.
- 1:30 to 3:00. Continuation of scientist reports
- 3:00 to 3:15. Break
- 3:15 to 4:00. Continuation of scientist reports
- 4:00. Adjourn
- 6:00 Meeting dinner. Cantonese Dinner
- Friday November 8
- 8:00 – 8:30. Breakfast buffet. Chefs Breakfast Table
- 8:30 to 11:00. Discussion, planning, comments from advisory board, recommendations for research, future meeting planning and more
- 11:00. End of meeting. Departures

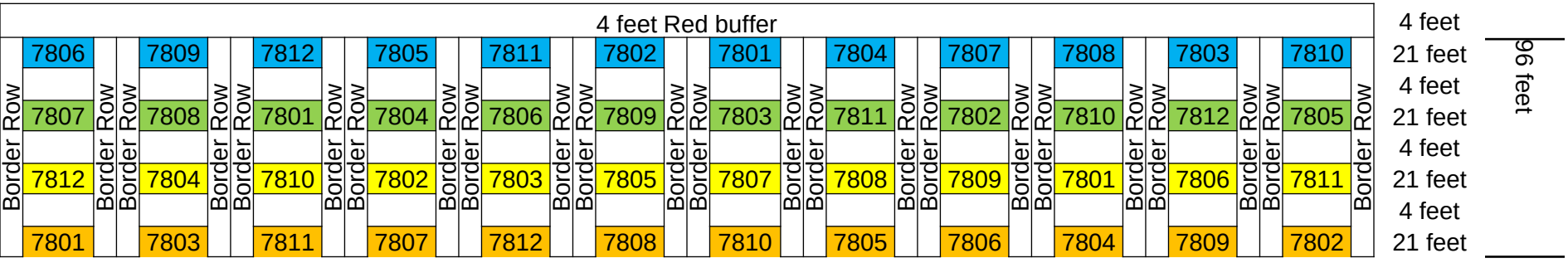
Is Dickeya spread by seed cutting?

- 2017 Field trials by Steven B. Johnson, UME, showed that Dickeya is not readily transmitted during seed potato handling and cutting operations
- NDSU trials: Dickeya inoculated seed mixed with healthy seed, handled, cut and planted separately
  - 2017 Plot trial in Florida
    - No stand loss, no spread to healthy seed
  - 2018 Commercial trial in Florida
    - 200 pounds seed inoculated with D. dianthicola mixed with 2500 pounds healthy seed and cut together and planted separately
    - No stand loss in healthy seed
    - > 90% stand loss in the inoculated seed
  - Some advisory Board members did not believe our data so we repeated the work in 2019

2019 trials

- Replicated trials in commercial fields in FL and MD
- 25 ft rows 4 reps
- 12 treatments using Dickeya free Atlantic seed
  - Mixed with seed naturally infected with D. dianthicola
    - Tubers tested positive for Dickeya by PCR
  - Mixed with seed infiltrated with D. dianthicola
  - Mixed with seed infiltrated with lab water/NB
  - Combinations of cut and whole seed
  - Infected seed removed R v W
  - No blackleg/seed decay observed either site
  - Cores and peels collected from 240 progeny tubers (5 tubers/trt/rep)
    - all tested negative for Dickeya by PCR
  - No differences in yield or grade

7801	Atlantic (100 whole seed)	Maine (11 whole seed)	Natural
7802	Atlantic (100 whole seed)	DRN (11 whole seed)	Lab Dickeya
7803	Atlantic (100 whole seed)	ATL (11 whole seed)	Lab water
7804	Atlantic (100 cut seed)	Maine (11 cut seed)	Natural
7805	Atlantic (100 cut seed)	DRN (11 cut seed)	Lab Dickeya
7806	Atlantic (100 cut seed)	ATL (11 cut seed)	Lab water
7807	Atlantic (100 whole seed)	Maine (11 cut seed)	Natural
7808	Atlantic (100 whole seed)	DRN (11 cut seed)	Lab Dickeya
7809	Atlantic (100 whole seed)	ATL (11 cut seed)	Lab water
7810	Atlantic (100 cut seed)	Maine (11 whole seed)	Natural
7811	Atlantic (100 cut seed)	DRN (11 whole seed)	Lab Dickeya
7812	Atlantic (100 cut seed)	ATL (11 whole seed)	Lab water



Multi-lab ring testing for Dickeya and Pecto

- Detection of Latent Dickeya in Potato Tubers : A simplified procedure
- Wash tubers to remove soil that may contain other Enterobacter that may cross-react in PCR testing
- Remove a stem-end core (approx. 0.5 in x 0.5 in/13 mm x 13 mm) and a stem-end peel strip (approx. 1.0 in/25 mm long) for individual tuber testing
- For seed lot testing, test 400 tubers consisting of 16 samples of 25 core and 25 peel samples comingled
- Wash the core and peel samples in tap water and a final rinse with distilled water. It is important to remove soil that may have Enterobacter spp. that could cause faint false positive bands after PCR
- Put each sample of 25 cores and peels in a re-sealable bag and add an equal wt/vol of quarter strength Ringer's solution
- Smash the sample, squeeze air from the bag and incubate at 30C for 24 hours
- Collect 2 ml supernatant liquid from each bag sample and centrifuge for 10 min at 14,000 rpm. Discard the supernatant liquid and use the pellet for DNA extraction
- Extract DNA using a Qiagen DNA extraction kit according to manufacturer's instructions
- Test for Dickeya by conventional PCR using pelADE or DiaC primers
- NOTE. For individual ring test tubers, each tuber should be tested individually

Dickeya/Pecto multi lab ring test results 6 Nov 19

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
Inoculated	Dd10^5	Dd 10^8	H_2O/NB	Pcc 10^5	Pcc 10^8	Dd 10^5 +Pcc 10^8
	Dd/Pcc	Dd/Pcc	Dd/Pcc	Dd/Pcc	Dd/Pcc	Dd/Pcc
Lab 1	1 0	1 0	0 0	0 1	0 1	1 1
Lab 2	1 1	1 1	0 1	0 1	0 1	1 1
Lab 3	1 0	1 0	1 0	0 1	0 1	1 1
Lab 4	1 0	1 0	1 0	0 1	0 1	1 1
Lab 5	1 0	1 0	0 0	0 1	0 1	1 1
Lab 6	1 0	1 0	0 0	0 1	0 1	1 1
Lab 7	1 1	1 1	0 1	0 1	0 1	1 1
Lab 8	1 0	1 0	0 0	0 1	0 1	1 1
Lab 9	1 0	1 0	0 0	0 0	0 1	1 0
Total	9 8	9 8	7 7	9 8	9 9	9 8

Notes and observations

- We did really well for detecting Dickeya, but not so well for Pecto
- No problem with detecting Dickeya
- Some difficulties detecting Pecto
  - False positives
  - Pecto ID – do we care?
  - Many primers available to use for Pecto; which is best?
- Can this simplified procedure can be recommended/used by private labs for Dickeya testing of seed lots
  - Nematode testing labs, soil labs, diagnostic labs, etc