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REBEL PIONEER CREATOR DEFENDER ADVENTURER EXPLORER

Soybean Cyst Nematode



REBEL PIONEER CREATOR DEFEND

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REBEL PIONEER CREATOR DEFENDER ADVENTURER EXPLORER

Presentation to
Getting it Right- Crop Production Meeting
Winkler, Manitoba
March 5, 2019



Important Tip

- Important stuff is indicated in Green
- All other stuff is optional, feel free to do something else
- Suggestions to do
 - Follow Mario on Twitter
 - Check what Trump has said today
 - Funny cat videos https://www.youtube.com/watch?v=hY7m5jjJ9mM

Table 1. Estimated soybean yield losses from diseases in the top 28 U.S. soybean-producing states and Ontario, Canada, in 2015.

Ontario, Canada, in 20	15.	
Disease/Pathogen	2015 Estimated Yield Losses for U.S. (thousands of bushels)	2015 Estimated Yield Losses for Ontario (thousands of bushels)
Root Rots and Seedling B	lights	
Soybean cyst nematode	109,288	3,696
Seedling diseases (caused by species of Fusarium, Pythium, or Rhizoctonia)	62,948	2,957
Root-knot nematode	12,366	0
Reniform nematode	4,438	0
Other nematodes (lesion, Columbia lance, sting, stubby root)	1,465	148
Leaf and Aboveground Di	seases	
Septoria brown spot	26,868	37
Frogeye leaf spot	17,662	15
Cercospora leaf blight	12,840	0
Downy mildew	4,383	7
Bacterial diseases (bacterial blight and bacterial pustule)	2,774	4
Virus Diseases (AMV, BPMV, SbDV, SMV, SVNV, TRSV, TSV)*	2,602	74
Other leaf and aboveground diseases (Phyllosticta leaf spot, target spot)	2,427	o
Purple seed stain	1,594	15
Rhizoctonia aerial blight	652	0
Soybean rust	157	0

2015 Disease Losses

Stem Diseases		
Sudden death syndrome	43,776	2,218
Sclerotinia stem rot (also known as white mold, caused by Sclerotinia sclerotiorum)	40,083	2,957
Phytophthora root and stem rot	28,275	1,479
Charcoal rot	20,808	15
Brown stem rot	17,389	74
Stem canker	12,349	222
Pod and stem blight	10,718	296
Anthracnose	5,188	0
Diaporthe/Phomopsis complex (seed rot)	3,612	44
Fusarium wilt and root rot	3,169	1,109
Other stem diseases (Phymatotrichopsis root rot, red crown rot, taproot decline)	2,253	o
Southern blight	523	0

^{*}AMV = alfalfa mosaic virus, BPMV = bean pod mottle virus, SbDV = soybean dwarf virus, SMV = soybean mosaic virus, TRSV = tobacco ringspot virus, TSV = tobacco streak virus.



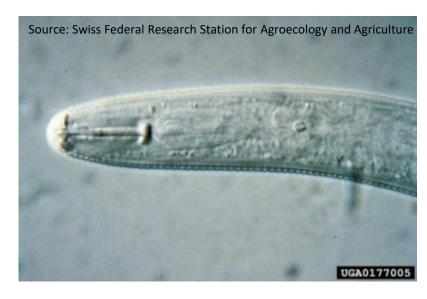
Soybean Cyst Nematode



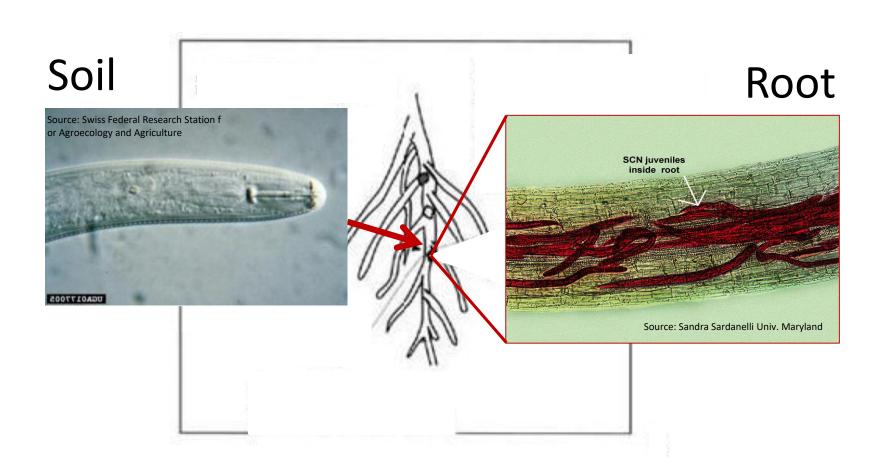
Soybean Cyst Nematode (SCN)

- Is a nematode (round worm) that parasitizes roots of soybean
- Like people, not all nematodes are bad, but SCN is bad

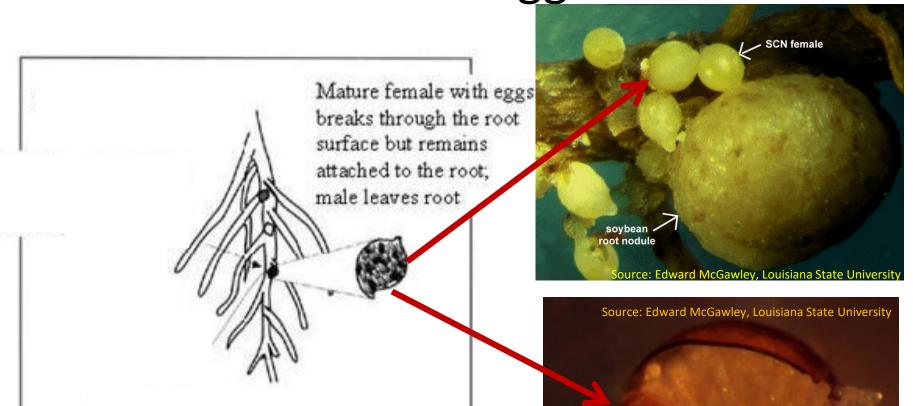




The Life of a SCN Female

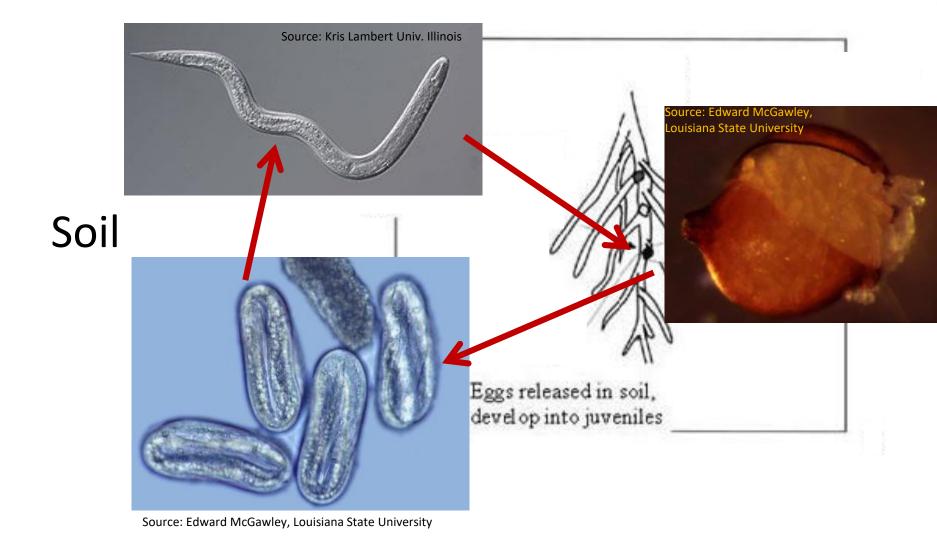


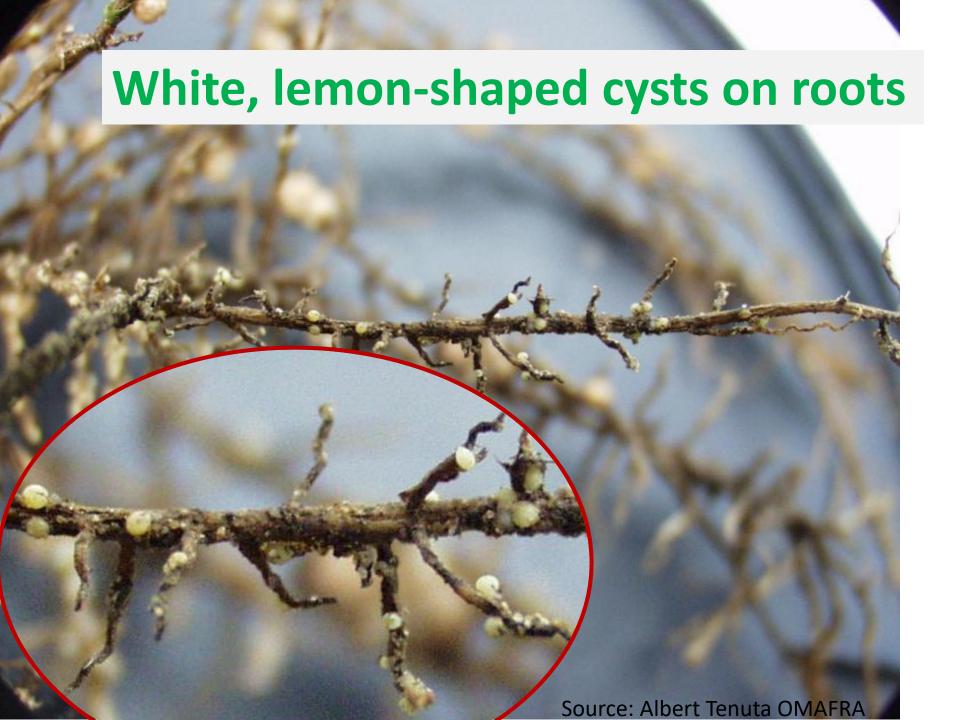
Female Settles Down to Feed and Produce Eggs

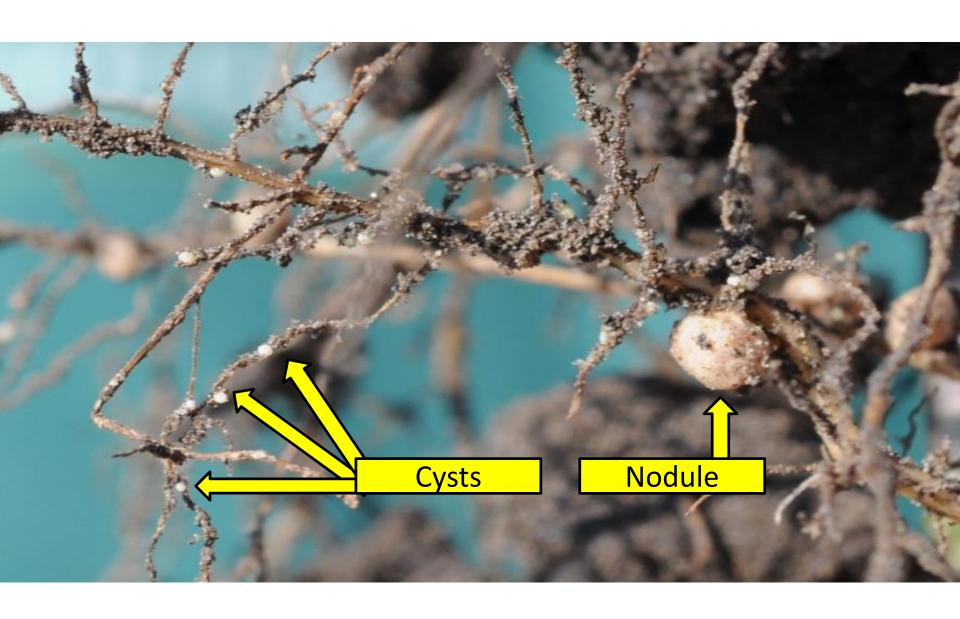


Source: Greg Tylka Iowa State Univ.

Female Becomes Cyst Eventually Rupturing and Releasing Eggs







Source: Albert Tenuta OMAFRA

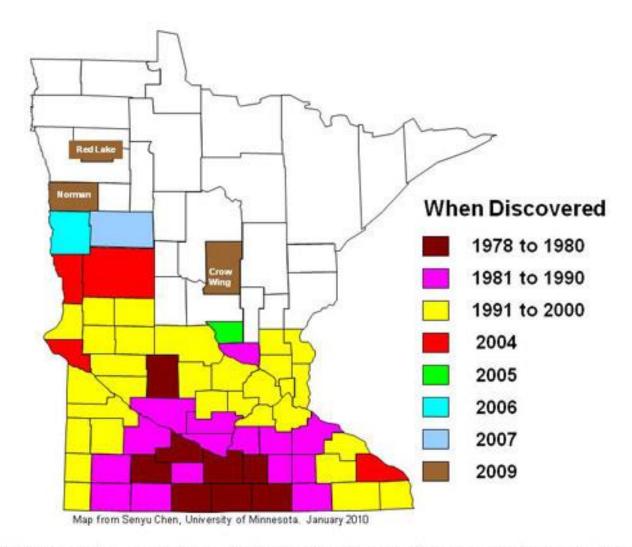
Damage Patches in Fields



SCN is Spreading to all Soybean Areas of Canada and U.S.

- Japan in 1880
- North Carolina in 1954
- Moved rapidly from there through much of soy growing area of the U.S.
- Minnesota in 1978
- Ontario in 1987
- North Dakota in 2003

SCN in Minnesota (2009)

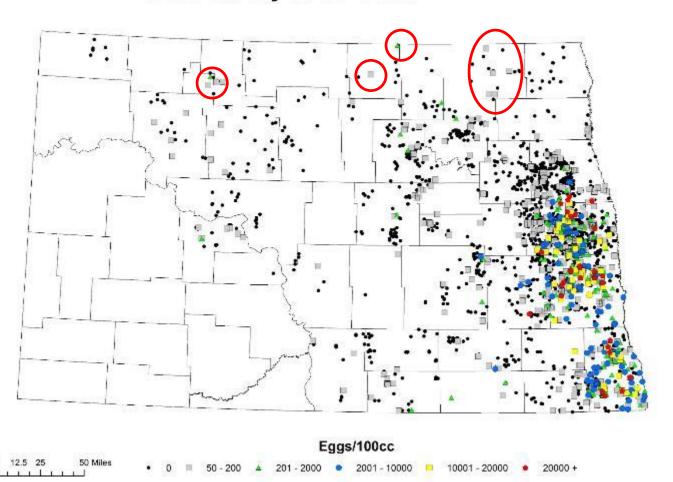


Minnesota counties infested with soybean cyst nematode

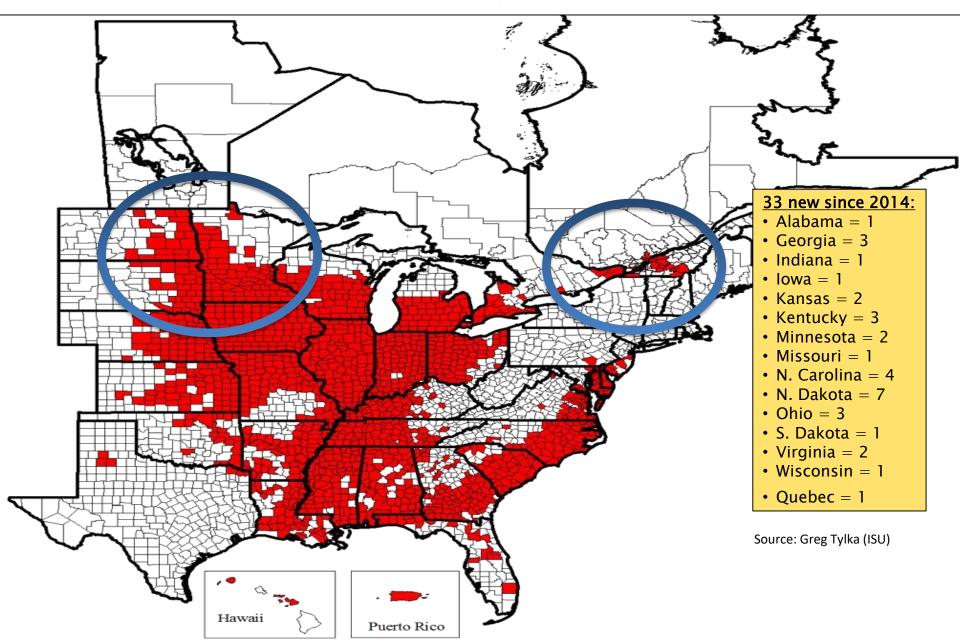
SCN Survey in North Dakota



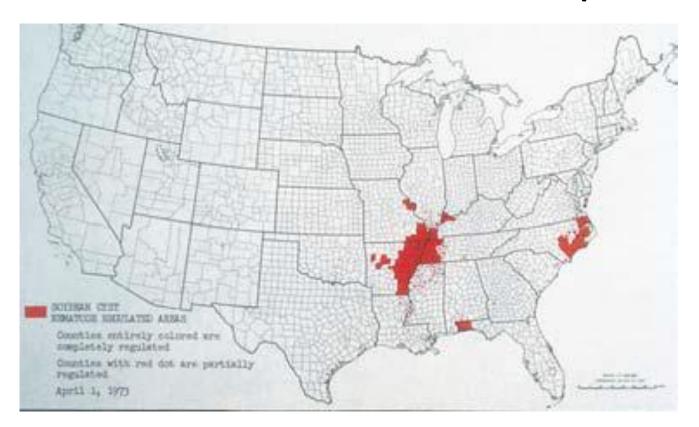
SCN Survey 2013 - 2017



Known SCN-Infested Counties - Feb. 2017



SCN in the U.S. (1973)



Spread of SCN Took U.S. Nematologists by Surprise

- Doesn't like cold soil (wrong)
- Doesn't like clay soil (wrong)

Can be Confused with Drown Outs





Effects of SCN on Soybean

What does it do?

- Takes away nutrients
- Water update disrupted
- Interferes with nodulation
- Damages roots (holes)

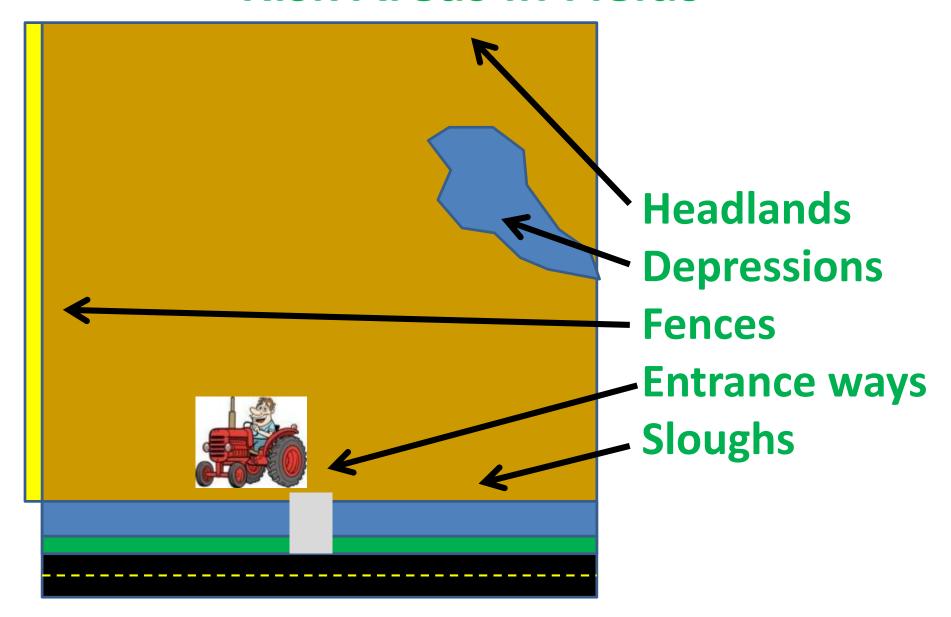
Field symptoms?

- Yellowed plants
 - Resembles Iron Chlorosis
- Stunted plants
 - Uneven height
- Early maturity
- Reduction of yield
- Fewer pods
- Damage shows earlier on sands

Avoid Host Plants in Fields

Crop Plants	Weed Plants
Adzuki Bean	American Vetch
Alsike Clover	Carolina Vetch
Bird's-foot Trefoil	Common Chickweed
Common Vetch	Common Mullein
Cowpea/Black-eyed Pea	Field Pennycress
Crimson Clover	Hemp Sesbania
Crownvetch	Henbit
Pinto, Navy, Cranberry, Black, Kidney,	Hop Clovers
Great Northern, Snap Bean	Milk Vetch
Hairy Vetch	Mouse-ear Chickweed
Lespedezas	Pokeweed
Lima Bean	Purple Deadnettle
Lupines	Purslane
Mung Bean	Shepherd's Purse
<u>Pea</u>	Wild Mustard
Soybean	Winged Pigweed
Sweet Clover	Wood Vetch

Risk Areas in Fields



Prevent Soil Movement Between Fields

- Purchase clean used equipment
- Wash implements and tires between fields
- Don't drive pickups between fields
- Clean footwear





Prevent Birds From Landing on Fields



Use Resistant Soy Varieties

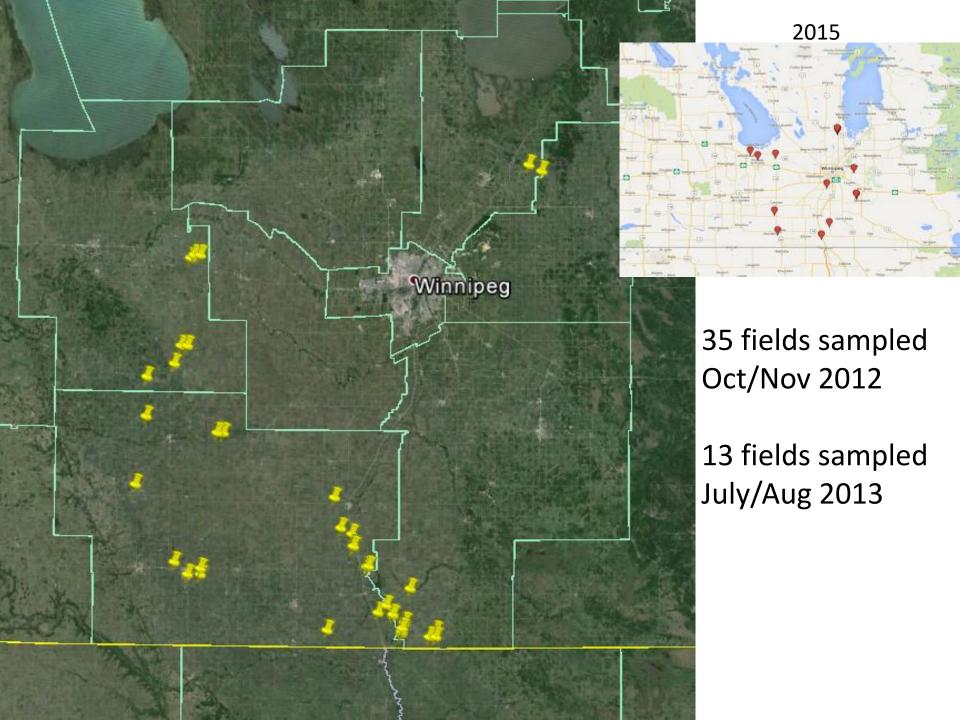


SCN in Manitoba?

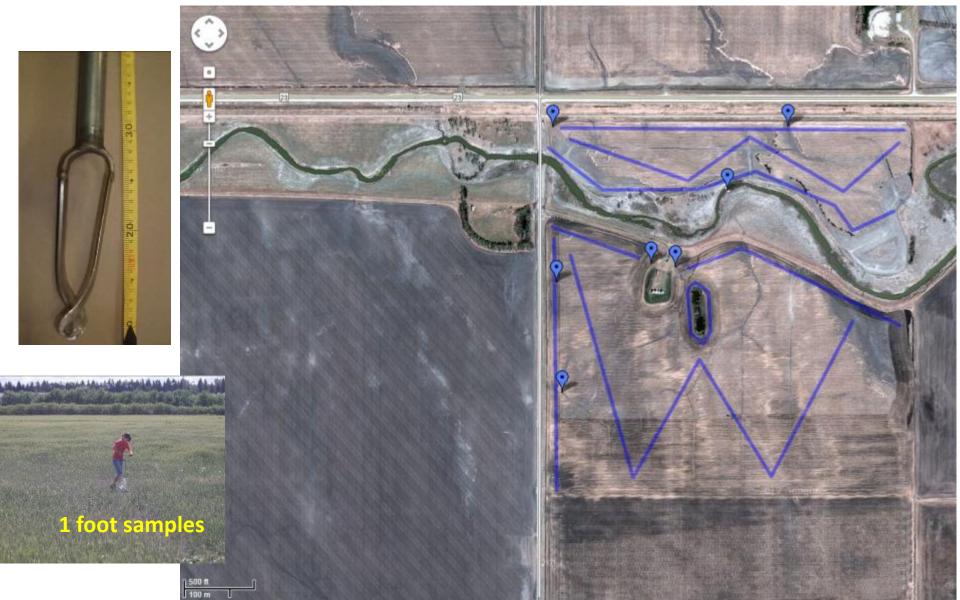
- Canadian Food Inspection Agency (CFIA) has done some survey work of random fields
- CFIA found in survey of potato soil in 2010 what seemed to be a lot of SCN in one field
- CFIA has removed SCN as a Regulated Pest in Canada and thus will not survey fields any longer

SCN Survey of Manitoba 2012-2015

- 76 soybean fields sampled
- > 5500 soil samples
- 487 composite samples for processing
- Priority fields based on
 - Proximity to water courses from U.S. that flood
 - Number of soybean years
 - History of dry beans
 - Sampled prone areas of fields

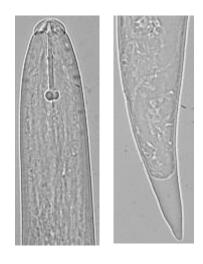


Collecting Soil Samples

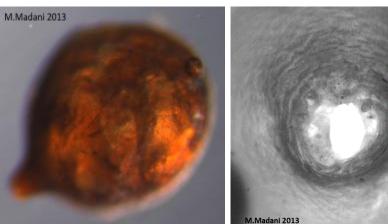


Juveniles and Cysts

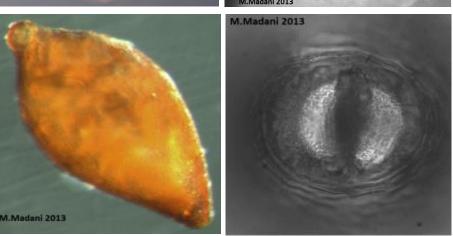




Circumfenestrate



Bifenestrate



Results 2012/13

- 37 composite samples from 22 fields had cysts
- Total of 60 cysts recovered
- 26 cysts were not damaged
- 23 cysts had circumfenestrate vulval cone structures – Cactodera, Punctodera, Betulodera
- 3 cysts were bifenestrate Heterodera

Results

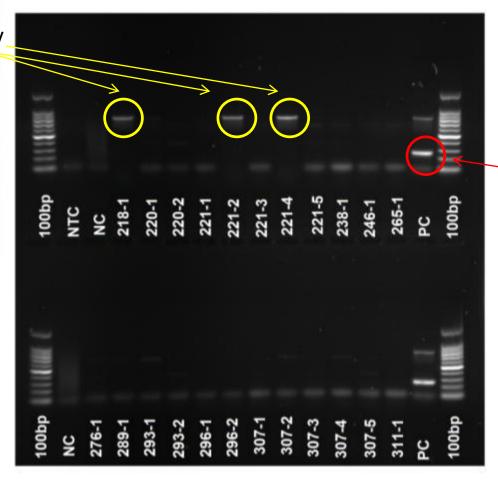
- 15 circumfenestrate cysts had eggs or juveniles
- 1 bifenestrate cyst had eggs and juveniles
- ITS sequencing, species-specific PCR
- Circumfenestrate cysts ITS matched Cactodera
- Bifenestrate cyst ambiguous Heterodera by morphology, SCN by 2/3 primer sets, Cactodera by ITS sequencing

Results 2014/15

- 28 fields sampled
- 205 composite samples analyzed
- 32 samples had cysts, but only a few each
- Most cysts were round and not lemon shaped
- Cone top patterns circumfenestrate
- 6 cysts yielded DNA for analysis, failed to be SCN

Species Specific PCR for SCN in 2015

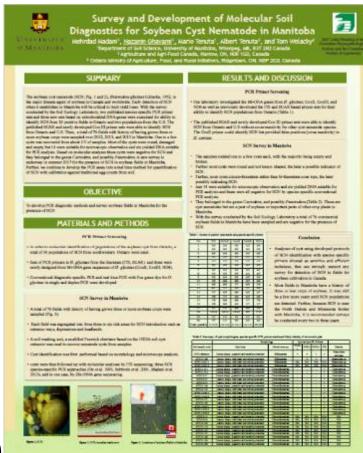
3 cysts yielding quality DNA but not positive For SCN



Positive control SCN yielding good DNA and giving band for SCN

SCN and Manitoba

- Cysts with quality for morphological and molecular analysis belonged to genera Punctodera and Cactodera
- Likely not of economic concern but on weeds
- But!!! Most of Manitoba's +1,200,000 acres of soybean acres is relatively new to production, thus over next 5-10 years likely establishment of SCN in Manitoba

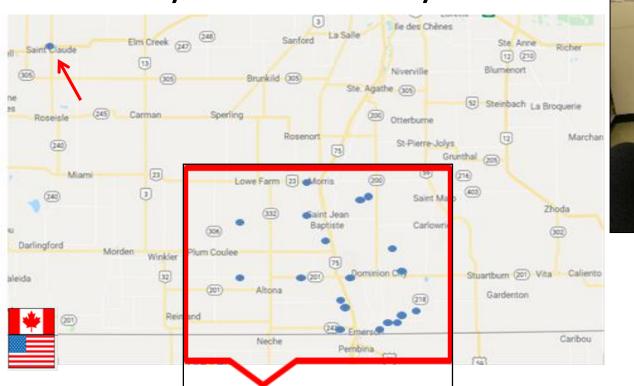


Manitoba 2017/2018 SCN Survey

Fall 2017, 29 soybean fields soil sampled

Samples have been extracted and being

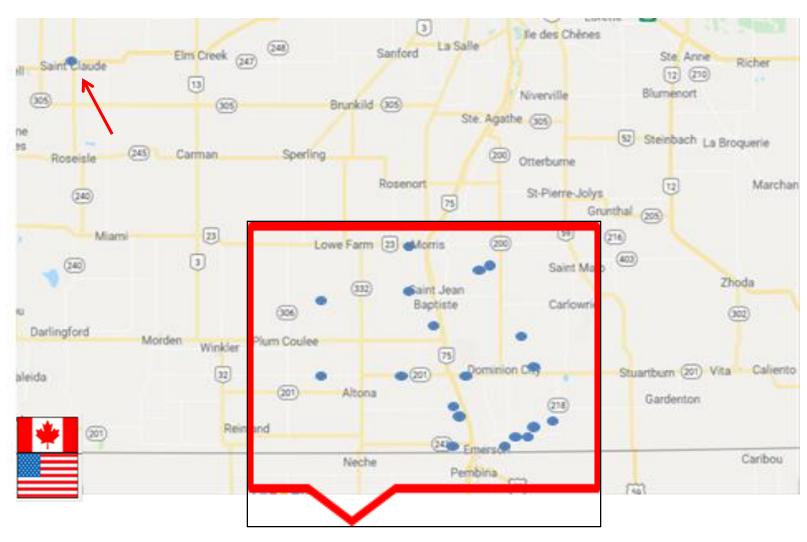
analyzed now for cysts



PhD student: Nazanin Ghavami

Soil Sampling

 29 commercial soybean fields along the Manitoba/U.S. border with history of soybean and edible bean cultivation and one field sampled by CFIA as part of nematode survey of seed potato farms which claimed there were lemon-shaped cyst in field, were sampled



Soil Preparation

- A total of 90 composite soil samples were obtained for about 3 samples for analysis per field
- Soil from each area was air-dried and 2.2 Kg soil kept in fridge for extracting cyst using a soil washing unit, a modified Fenwick elutriator based on the USDA soil cyst extractor









Cyst Extractor

- Modified Fenwick elutriator based on the USDA soil cyst extractor
- We got an efficiency of 70 % in recovery of cysts from reference samples









Ethanol Floatation

- Collected debris from cyst extractor was dried and then floated in ethanol and trapped onto filter paper
- The ethanol flotation step had an efficiency of 95%

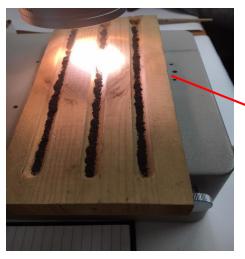




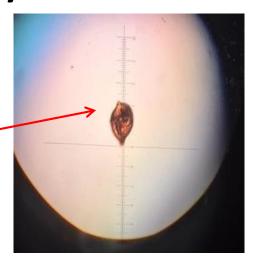




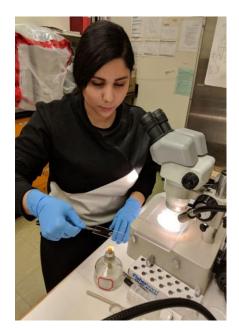
Cyst Identification Based on Morphology









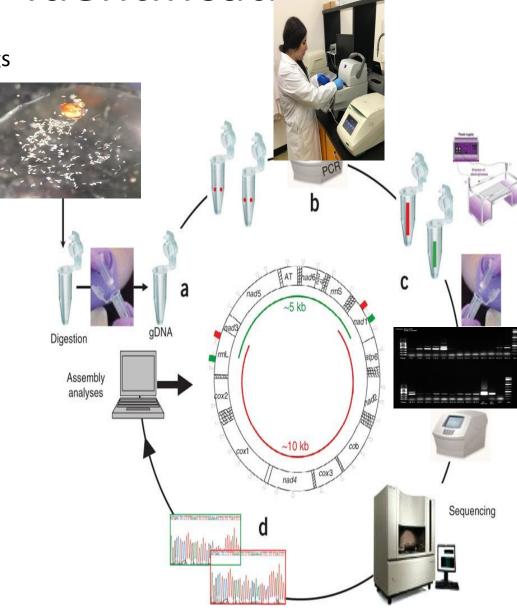




Molecular Identification

✓ DNA extraction from juveniles and eggs suspension by Crushing Method with glass Beads

- Quantifying DNA extracted Using Nanodrop Spectrophotometer
- ✓ DNA Amplification Using Polymerase Chain Reaction using species specific primers (Conventional)
- ✓ Visualizing DNA
- ✓ DNA Purification
- Sequencing based on the NCBI database.



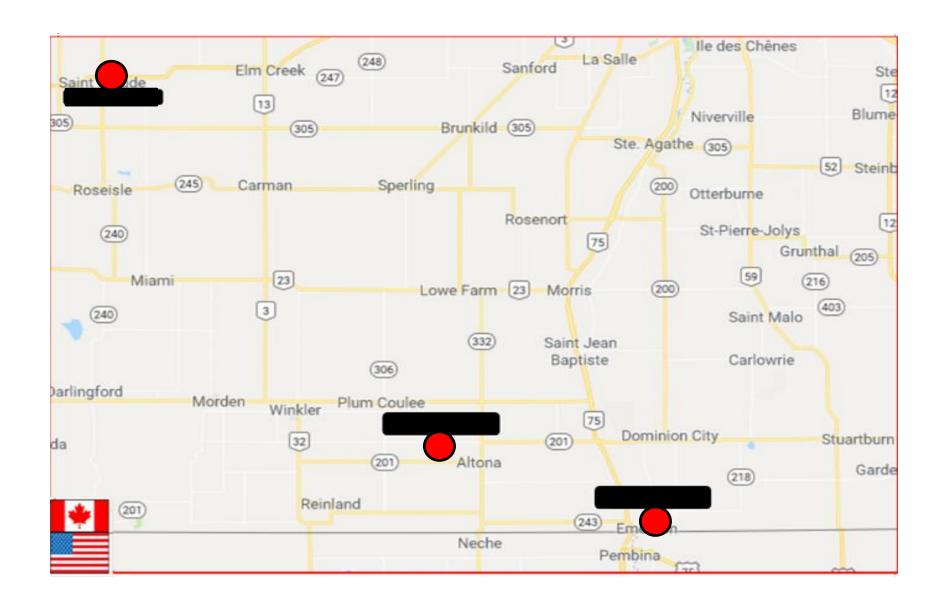
Preliminary Results 2018

- √ 30 commercial soybean fields were sampled
- ✓ A total of 90 composite soil samples were obtained
- ✓ Overall, 17 of the composite samples from 12 fields had nematode cysts
- ✓ One to a few cysts were recovered from each of these 17 composite samples.
- ✓ In total, 42 cysts were recovered and 30 of the cysts from seven fields were brown and lemonshaped as expected of SCN

Preliminary Results

- √ 10 lemon-shaped cysts and bifenestrate were obtained from three fields
- ✓ PCR of 14 lemon-shaped cysts the 252 bp (CoxIIIF1- CoxIIIR1) and 477 bp (SCNFI-SCNRI) genes regions were SCN
- ✓ Based on the morphological characters and PCR assays conducted so far, we speculate that three fields had 2, 14 and 4 SCN cysts / 5 lbs soil
- ✓ DNA sequencing of multiple regions is being done

Suspected Fields



On-going

- ✓ DNA sequencing of multiple regions for the SCN cysts is now being done
- ✓ The positive 3 fields will be re-sampled in spring 2019
- ✓ Soil from the 3 fields will be planted to soybean in the greenhouse for development of cysts on roots

SCN Emerging Issue for Dry Beans

plant disease

Editor-in-Chief: Alison E. Robertson
Published by The American Phytopathological Society

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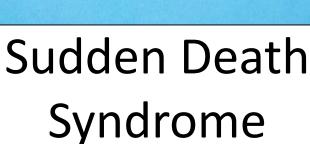
DISEASE NOTES

First Report of the Soybean Cyst Nematode
Heterodera glycines Infecting Dry Bean
(Phaseolus vulgaris L.) in a Commercial Field
in Minnesota

G. P. Yan, A. Plaisance, I. Chowdhury, R. Baidoo, A. Upadhaya, J. Pasche, S. Markell, and **B. Nelson**, North Dakota State University, Department of Plant Pathology, Fargo 58108-6050; and **S. Chen**, University of Minnesota, Department of Plant Pathology, St. Paul 55108.

- 2016 stunted patches in dark-red kidney bean field
- Roots infested with SCN females
- Soybean last grown in 2010





- First reported 1971 in Arkansas
- Disease complex of SCN with Fusarium virguliforme
- Occurs after first flowering
- In Minnesota and South Dakota



Scout for SCN

- Fields more than 3 years of soybean
- Get out of the truck and walk
- 30-45 days after emergence, gently lift roots with spade, dunk in bucket of water, look for females using a hand lens
- Collect soil samples and SCN test (Agvise or Soil Ecology Lab U Manitoba)







How to Check Roots

http://www.nwroc.umn.ed u/Cropping_Issues/2010/Ju ly_20/SoybeanCystNemato deScouting/index.htm

Soil Sampling for SCN

- Every third soybean crop year
- Sampling in fall following crop harvest and before soil freezes
- Following soybean harvest, sample directly within harvested rows before tillage
- Following other crops, sample after fall tillage, if you till
- Sample top eight inches
- Use a soil push probe or small diameter soil auger
- Take 15 cores for a sample from every 20 acres
- Sample specifically for trouble soybean area

More on Soil Sampling

- Put cores into a bucket
- Mix the cores and place into a ziplock freezer bag
- Label both sides of bag with marker for name, legal, date, field sample number
- Keep bag out of sun
- Place in refrigerator
- Drop off samples to Agvise as you would do for soil fertility testing
- SCN is not a regulated pest, so call Mario if samples come back positive

Exam Study Points

- SCN can silently rob yield
- SCN is confused with other crop problems
- SCN is marching in our direction and is inevitable
- Can delay and lessen damage by
 - Clean machinery, tires and footwear
 - Know your field risk areas
 - Dig plants to scout fields
 - Weed suppression
 - Avoid tight rotations
 - Don't rotate with edible beans and pea
 - SCN soil test
 - Use resistant varieties
 - Bird suppression
- Scout roots for cysts, and soil sample every 3rd soy crop
- You need to know everything for the exam!

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Acknowledgements

Numerous colleagues for pictures

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Canada Research Chair Program in Applied Soil Ecology









University
of Manitoba

Three Things Are Forever

- Diamonds
- Taxes
- SCN

