



Clubroot in Canola- Research Updates of 2020

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Canola Expo 2020
December 8th, 2020**

Clubroot



- Causal agent *Plasmodiophora brassicae*
- Obligate biotrophic soil-borne plant pathogen
- Not a fungus/amoeba/slime mold but has some characters similar from each
- Infects hosts of brassica family
 - E.g. Canola, cauliflower, cabbage, rutabaga, radish, turnip, brussel sprouts, kale etc.
 - Susceptible brassica weeds: wild mustard, Shepard's purse, volunteer canola, stink weed
 - Model Organism: *Arabidopsis*
- Prefers acidic soils but found in the soils of pH up to 7.2
- Once in the soil can live as resting spores up to 20 years
- Pathogen infects roots; causes galls there by restricting the flow of water and nutrients to the plant
- If 100% of plants infected results in 50-80% reduction in yields (Europe and Sweden Research)
- Seen 25% of yield losses in Cavalier County, ND

Best Management Practices



To manage clubroot on canola in
North Dakota
Current Research is focused on Best
Management Practices

Source: Canola Council of Canada

Research Objectives studied in 2020

- **Statewide Clubroot Survey:**
 - Visual
 - Identification and Quantification of clubroot resting spores from soil
- **Clubroot management studies**
 - Seed treatments
 - Germplasm Evaluation
 - Canola Varietal Evaluation
 - Efficacy of Surfactants to manage clubroot
 - Efficacy of Surfactants with lime and without lime
 - Dose/rate determination of lime
- ***P. brassicae* Pathotypes of North Dakota**



Statewide Clubroot Survey

– Visual

- Walking in a W pattern and uprooting the stubbles and look for presence of galls

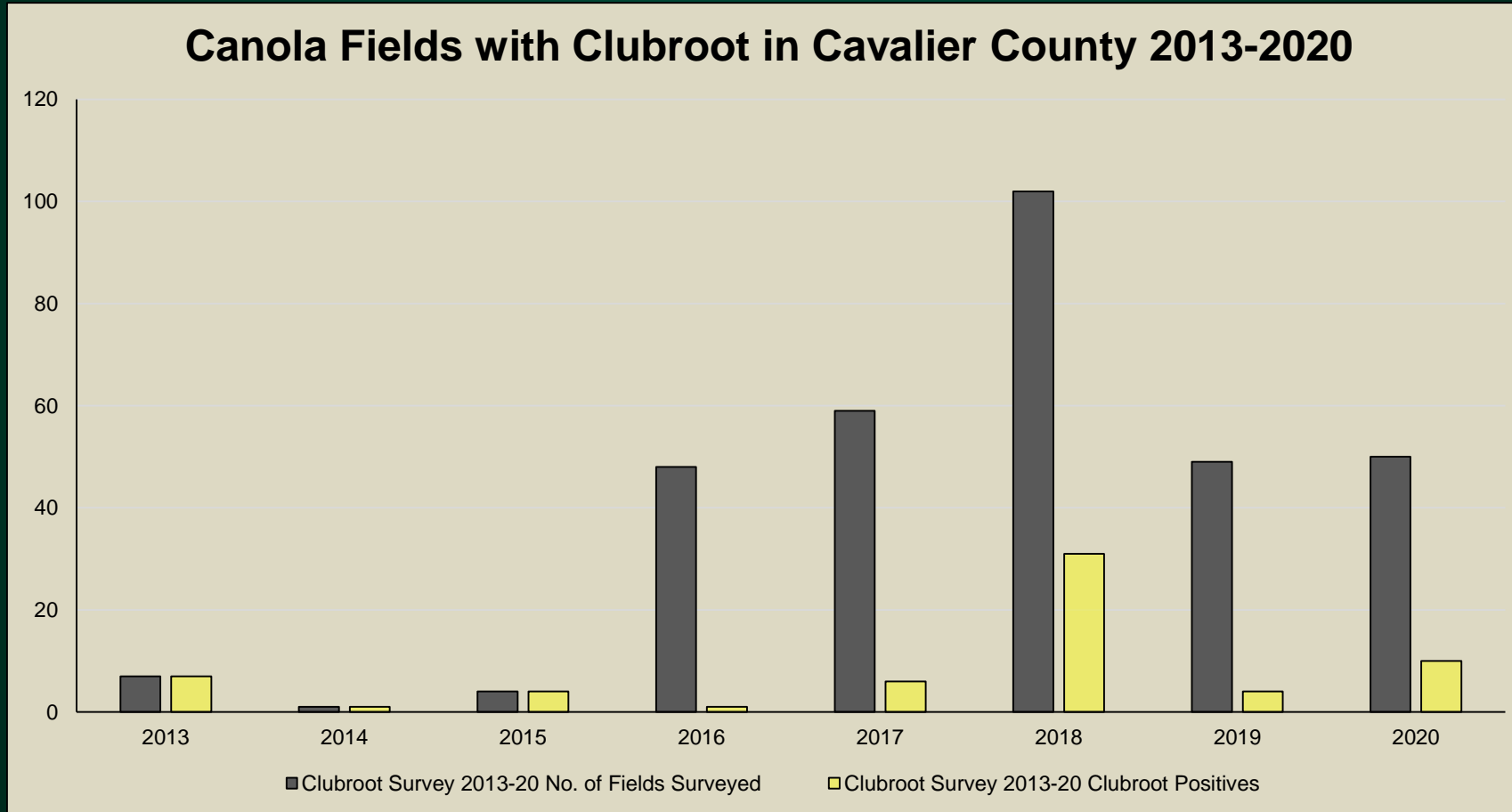


– Identification and Quantification of clubroot resting spores from soil

- Used Q-PCR the advanced Molecular technique to quantify the clubroot resting spores in soil
- This enables us to identify fields infected with clubroot if we missed seeing symptoms during standing crop
- Also determines the number of spores present per gram of soil

Clubroot on Canola in Cavalier County:2013-2020

- 20% of the fields found with clubroot (Visual galls)

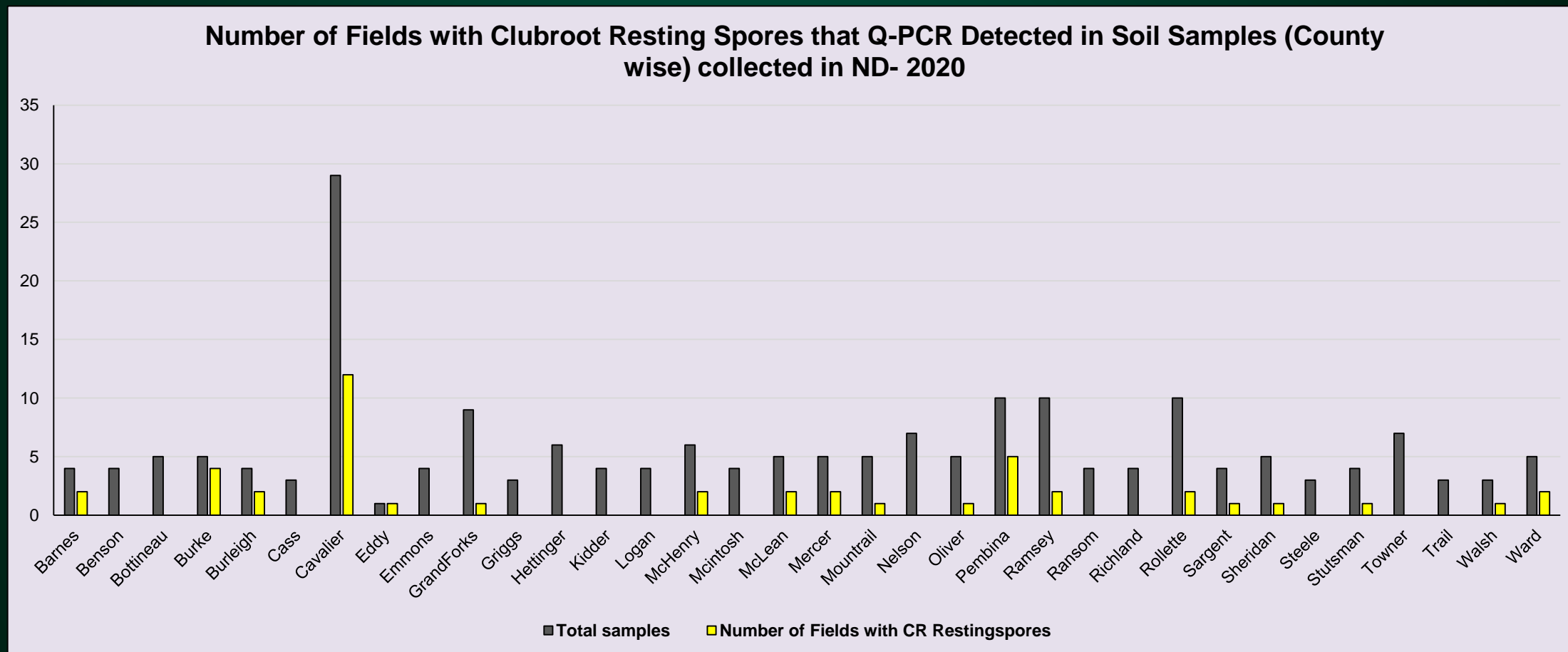


Clubroot Survey in Various Counties of North Dakota

- Visual galls found on brassica vegetables in McHenry County
- Resting spores found in soils-Confirmed with molecular tests using Q-PCR



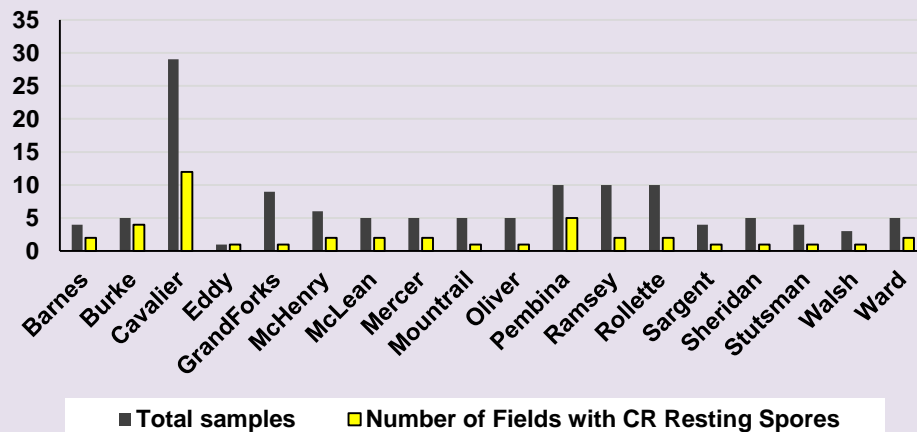
Clubroot Resting Spores found in soil samples from various Counties in ND



Fields with Clubroot Resting Spores found in Soil Samples from various Counties in ND-2020

Q-PCR Assays detected 18 out of 34 Counties had fields with Clubroot Resting Spores

Counties with Clubroot Resting spores in the soil samples-2020



Quantified resting spores of *P. brassicae* from all those samples ranged from 500 to 40 million spores per gram of soil (minimum detection limit of the assay being 10 resting spores/gm of soil).

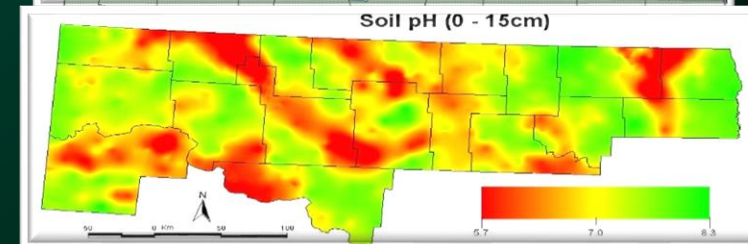
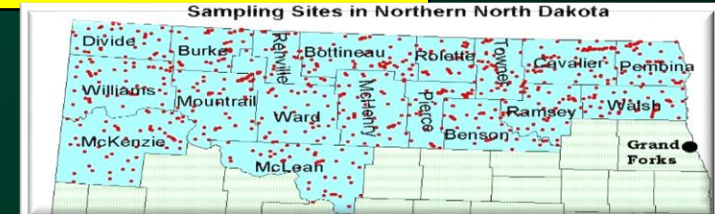
Number	County Name	Total samples	Number of Fields with CR Resting Spores
1	Barnes	4	2
2	Burke	5	4
3	Cavalier	29	12
4	Eddy	1	1
5	GrandForks	9	1
6	McHenry	6	2
7	McLean	5	2
8	Mercer	5	2
9	Mountrail	5	1
10	Oliver	5	1
11	Pembina	10	5
12	Ramsey	10	2
13	Rollette	10	2
14	Sargent	4	1
15	Sheridan	5	1
16	Stutsman	4	1
17	Walsh	3	1
18	Ward	5	2

Interpretation of State wide Soil Sample Quantification Tests

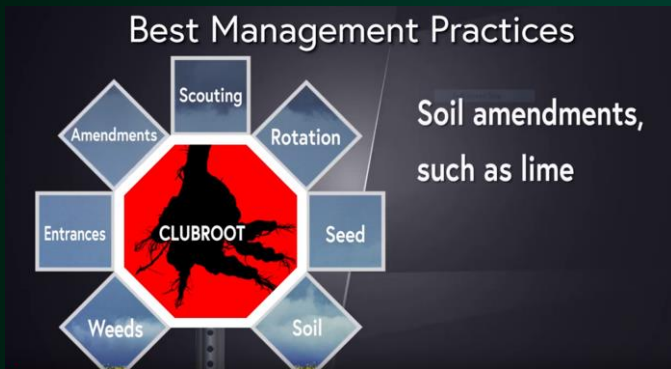
County	Location	Spores/Gram	Crop	GPS Coordinates	pH
Barnes	Leal	500	Soybean	47.103884; -98.311069	7
Stutsman	Wadsworth (N.O.)	3 million	Pasture	47.229373; -99.332308	8
Barnes	Binghamptom	5 million	Canola	46.753053; -97.802422	7.5
Burleigh	Gibbs	90000	Pasture	46.852463; -100.66080	6.6
Burleigh	Driscoll	50000	Wheat	46.839536; -100.145555	6.8
Sargent		7000	Pasture	46.060146; -97.491226	7.9
Cavalier	Hay township	40 million	Canola	Anonymous	5.6
Eddy	EC20-1	1000000	Wheat	47.81445; -99.14765	7.5
Grand Forks	GFC20-7	500000	Cover Crop	47.80447; 97.55955	6.9
Rolette	RC20-8	700000	Canola	48.59576; -99.71068	5.8
Rolette	RC20-6	360000	Canola	48.67525; -99.52151	6.5

Samples form fields and counties represented with yellow background are to be monitored closely

Waiting for complete pH results



CLUBROOT MANAGEMENT STRATEGIES



Objectives Tested

- **Clubroot management studies**
 - Germplasm Evaluation
 - Canola Varietal Evaluation
 - Seed treatments
 - Soil amendments with lime and without lime
 - Dosage/Rate determination of lime
 - Efficacy of Surfactants to manage clubroot
- **On going *P. brassicae* Pathotype Study with Canadian Plant Pathologists**

Land Preparation



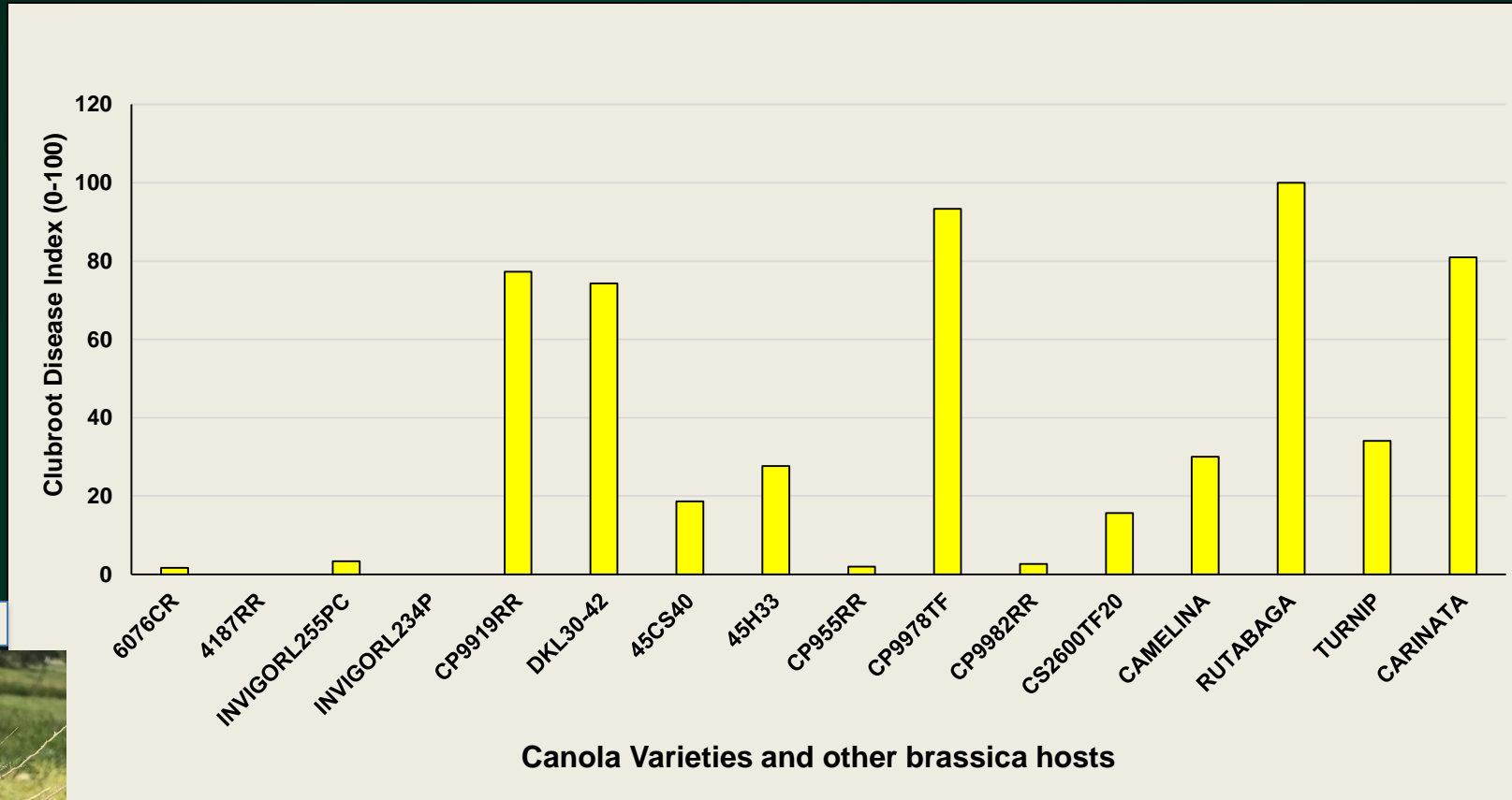
Objective 1: Canola Cultivar Evaluation along with other Brassica Hosts to Clubroot-2020

Aim: To evaluate performance and to monitor resistance breakdown

Cultivar	Description
6076CR	Brett Young Seeds
4187RR	Brett Young Seeds
INVIGOR L255PC	BASF
INVIGOR L234P	BASF
CP9919RR	Croplan Genetics
DKL30-42	Cargill
45CS40	Pioneer (Corteva)
45H33	Pioneer (Corteva)
CP955RR	Croplan Genetics
CP9978TF	Croplan Genetics
CP9982RR	Croplan Genetics
CS2600TFR	Canterra Seeds
Camelina	Winter Variety 'Joelle'
Rutabaga	Variety 'Laurentian'
Carinata	Unknown Variety
Turnip	Variety 'Purple Top White Globe'

16 treatments
4 replications
Randomized Complete
Block Design

Canola Cultivar Evaluation along with other Brassica Hosts to Clubroot-2020



Clubroot galls on Carinata

Resistant Cultivar



LSD: 31.15
P-Value: 0.00001*

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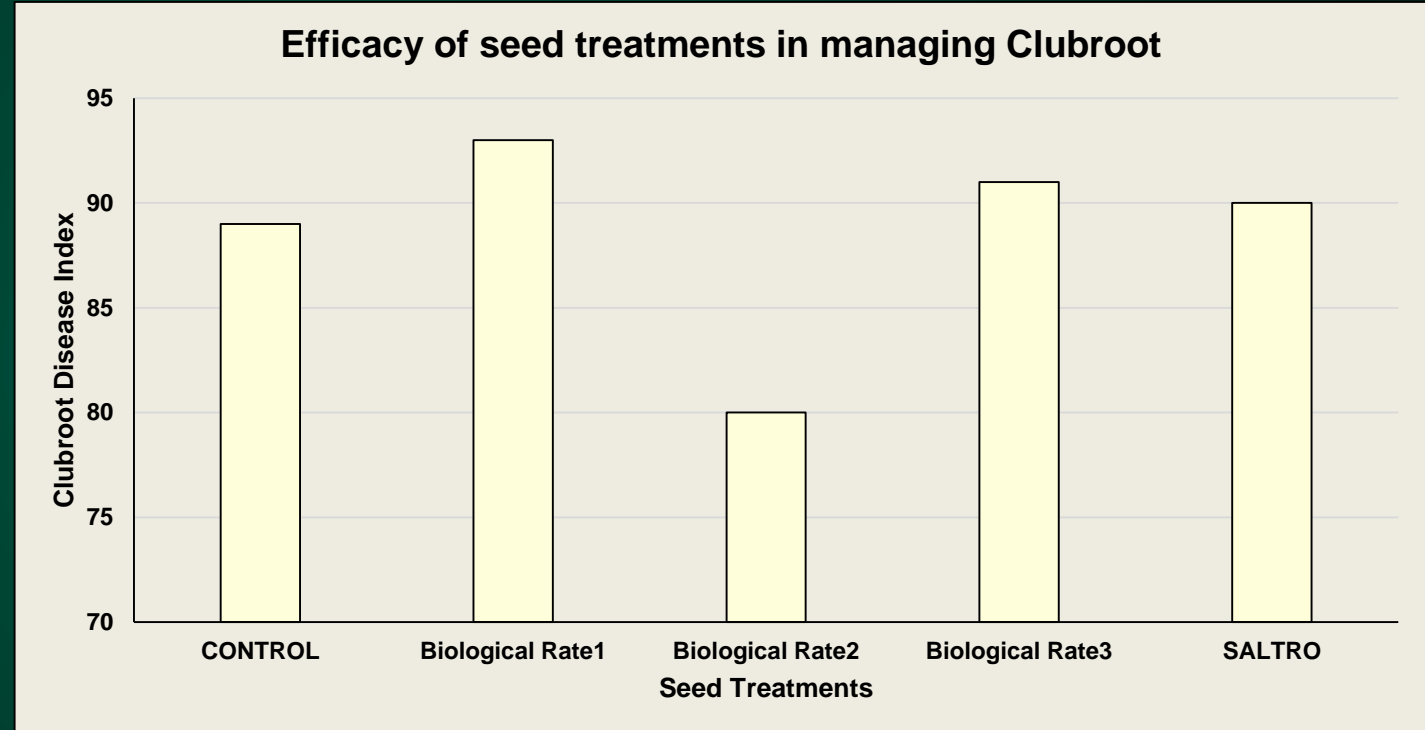
Scale:

DI <30% Resistant
DI 30-69% Intermediate
DI > 70 Susceptible
Validity of Trial >60% DI in susceptible check

Objective 2: Evaluation of Seed treatments

Tested on: cv. Westar
5 treatments
4 replications
Arranged in
Randomized Complete
Block Design
Evaluated after 60days

*None of the seed
treatments tested had
effect on clubroot
control



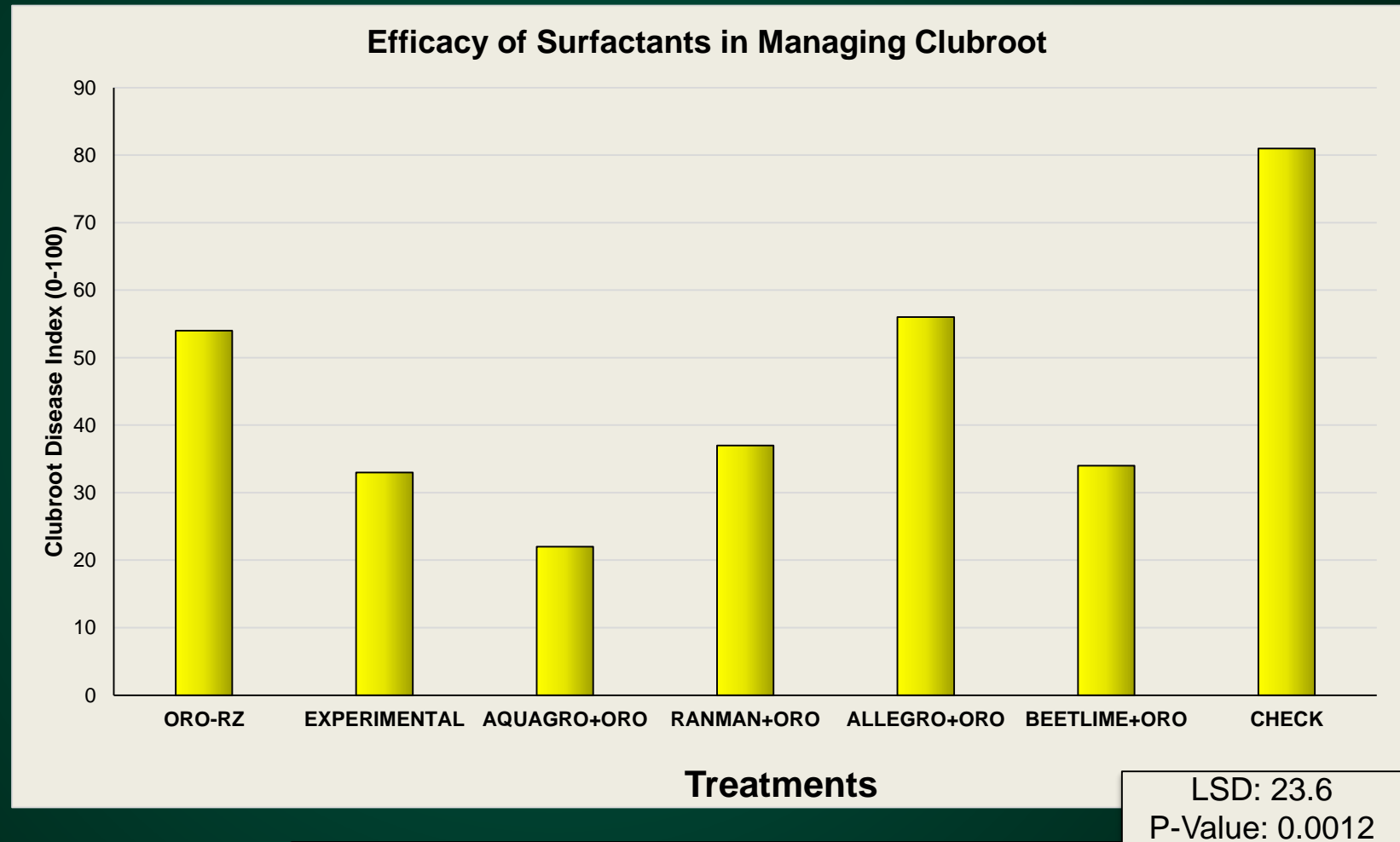
LSD: 13.95
P-Value: 0.35

Objective 3: Evaluation of Surfactants



Evaluation of Surfactants

Tested on: cv. L233P
7 treatments
4 replications
Arranged in Randomized
Complete Block Design
Evaluated after 60days



Objective 4: Evaluation of Surfactants with and without lime



Soil sample collection before application and after application of soil amendments



Soil Drenching of liquid Surfactants formulations and Chemicals

Evaluation of Surfactants with and without lime

cv. L233P

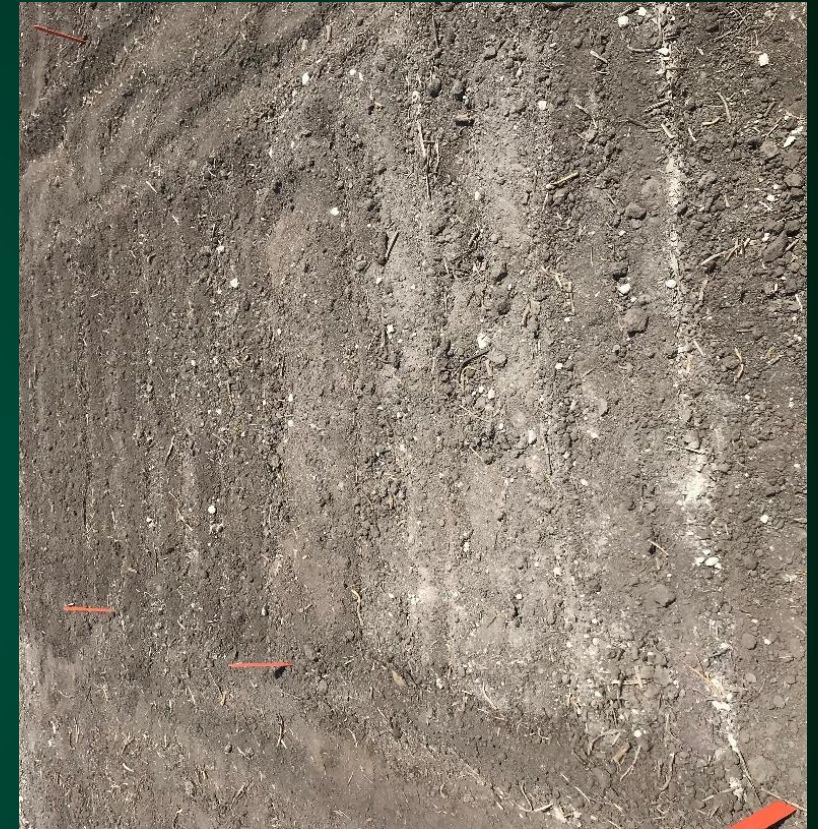
	Clubroot Disease Index
	P-Value
Bloc	0.479
Main Plot (Lime vs without Lime)	0.3275
Main Plot*Bloc	0.9504
Sub Plots	0.0001
Main Plot*Sub Plot	0.5752

Treatments	Rate	CR DI
Ranman+ORO	20 fl oz+2 pt/A	20
ORO Zero	CHK	84
ORO79 TWO	2 pt/A	33.5
ORO79 FOUR	4 pt/A	16
ORO79 EIGHT	6 pt/A	23
ORO09	4 pt/A	21.5
Mean		32.9
CV%		65
LSD(0.05)		22
p- Value (0.05)		0.00001

*The interaction results indicate that there were no differences among the treatments under the influence of lime applied and non-lime applied blocks

*There were differences in pH before and after application treatments in lime and surfactants applied blocks but not in surfactant alone applied blocks

Objective 5: Evaluation of soil amendments at various Doses/rates to manage clubroot



Lime applied plots

Soil amendments Dose/rate Response in Managing Clubroot

cv. L233P

Source	P-value	Fac_A	CR DI	Fac_B	CR DI
Bloc	0.7299	Beet lime	31	ZERO	81
Fac_A (Treatments)	0.0469	Pellet lime	48	5 t/ha	35
Fac_B (Rates)	0.0006	Wood Ash	57	10 t/ha	37
Fac_A*B	0.293	Mean	45	15 t/ha	29
		CV (%)	67	Mean	45
		LSD (0.05)	22	CV (%)	67
		p-Value (0.05)	0.0469	LSD (0.05)	25
				p-Value (0.005)	0.0006

pH results: There were significant differences among the treatments tested in terms of increase in pH from before application to after application evaluation

Pathotypes of *Plasmodiophora brassicae* present in North Dakota

Sample	North Dakota clubroot Pathotype Designation		
	Some et al. (1996)	Williams (1966)	Canadian Clubroot Differential Set
FFCR	P3	8	Novel
MMCR	P3	2	2C
PBCR-2	P2	8	8N
RBCR-4	P3	8	Novel
RBCR-5	P3	8	8D
YCR-16	P3	8	Novel

- The *P. brassicae* pathotype composition in North Dakota was quite distinct from that reported previously from Alberta, Canada, where the clubroot outbreak is most severe.
- None of the pathotypes identified could overcome first generation resistance, and
- In North Dakota, clubroot may still be managed by planting CR canola in a minimum 3-year rotation.

Lime application on germplasm evaluation studies of Canola diseases blackleg and white mold





Literature available on clubroot from NDSU



Chapara V, et al., J. Agron Agr Sci 2019, 2: 208

HSOA Journal of Agronomy and Agricultural Science

Short Communication

Prevalence of Clubroot on Canola in North Dakota

Chapara V¹, Kalluri N¹, Lubnow N¹ and Chirumamilla A¹
¹Langdon Research Extension Center, North Dakota State University, Langdon, USA
 NDSU Extension Service, Langdon, ND-58249, USA

Abstract

Clubroot of canola was regular and more prevalent than reported in Cavalier County in the current survey. Prevalence of clubroot on canola has been increasing at rapid pace in North Dakota and has been confirmed in 33 fields in Cavalier County in 2018, which is the first time more than 10 fields in 2017. Clubroot is a soil-borne disease that causes swelling and galls on the plant roots of Brassica family (Dolan 2009). These galls cause reduced premature root growth of canola plants and yield loss in the canola (Strelkov et al. 2005). Characterization of roots for galls by digging up from the canola plants is the quick identification of clubroot incidence (Chapara and Wright 2012). Clubroot disease incidence and development is favored by acidic soils (Strelkov et al. 2005). However, later research proved that clubroot on canola is not only limited to acidic soils but can also occur in alkaline soils (Strelkov et al. 2007). The highest degree of clubroot disease incidence was observed at pH 6.6 (Palm 1963). *P. brassicae* soil-borne obligate parasite, thrives in the soil as resistant spores that can remain viable in the soil up to 17.3 years indicating

Clubroot of canola caused by *Plasmodiophora brassicae* (Woronin) is the new emerging disease in North Dakota state as identified in canola in 2013 (Chapara et al. 2014). Clubroot is a soil-borne disease that causes swelling, or galls on the plant roots of Brassica family (Dolan 2009). These galls cause reduced premature root growth of canola plants and yield loss in the canola (Strelkov et al. 2005). Characterization of roots for galls by digging up from the canola plants is the quick identification of clubroot incidence (Chapara and Wright 2012). Clubroot disease incidence and development is favored by acidic soils (Strelkov et al. 2005). However, later research proved that clubroot on canola is not only limited to acidic soils but can also occur in alkaline soils (Strelkov et al. 2007). The highest degree of clubroot disease incidence was observed at pH 6.6 (Palm 1963). *P. brassicae* soil-borne obligate parasite, thrives in the soil as resistant spores that can remain viable in the soil up to 17.3 years indicating

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 Chapter V, Kalluri N, Lubnow N, Chirumamilla A (2019) Prevalence of Clubroot on Canola in North Dakota. J. Agron Agr Sci 2019, 2: 208. Received: February 11, 2019; Accepted: February 14, 2019; Published March 01, 2019

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Soil Sampling to Quantify Clubroot Spores From Soil in North Dakota

Note: Please complete this form in its entirety to identify the soil sample location. The information will be used to compile a distribution map of clubroot infestations in North Dakota.

County: _____ Grower's name: _____ (will remain confidential) Phone: _____

Field ID	Latitude N	Longitude W (-)	Field Identity			
			CR	Township	Range	Section

Latitude and longitude in decimal degrees is preferred.

Example: N: 48.659444 W: 98.24444 (GPS coordinates) - GPS coordinates can be obtained through "preferences" or other "setup" functions within the systems software.

Soil Sampling Survey Procedure

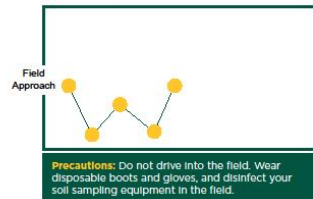
1. Walk in a "W" pattern in the field starting from the main entrance of the field (avoiding low spots or flooded areas, low pH spots and areas of diseased patches as shown in the figure below).
2. At each sample point, clear away residue on the soil surface and collect soil core or scoop from the top 3 to 6 inches (a representative sample is five scoops or cores from each field). Be sure to maintain 300 feet from point to point of soil collection.
3. Air-dry the soil samples indoors in paper boxes and send or drop them off at one of the following address:

Zhaohui Lu
 Department of Plant Pathology
 NDSU Department 7660
 P.O. Box 6050
 Fargo, ND 58108-6050

UPS/FEDEX to Zhaohui Lu
 Department of Plant Pathology
 Walster Hall 306
 Fargo, ND 58102

Venkat Chapara
 Langdon Research Extension Center
 9280 107th Ave. N.E.
 Langdon, ND 58249

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 North Dakota State University, Fargo, North Dakota



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Canola Diseases: Clubroot

Medicine Smith, NDSU Extension Agent, Pembina County
 Anita Chirumamilla, NDSU Extension Agent, Cavalier County
 Lindy Berg, NDSU Extension Agent, Towner County
 Venkata Chapara, Plant Pathologist, Langdon Research Extension Center
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 Luis E. del Rio Mendoza, Canola Pathologist, Department of Plant Pathology
 Sam Markell, Extension Plant Pathologist, Department of Plant Pathology

Clubroot is a serious disease threat to canola

product the disc County of North are been North D Once is elimin detectio manage and limit

Causes

The path brassicae kingdom groups P. brassicae such as canola, and weevil western pathotype composition in North Dakota was quite distinct from that reported previously from Alberta, Canada, where the clubroot outbreak is most severe. None of the pathotypes identified could overcome first generation resistance, and in North Dakota, clubroot may still be managed by planting CR canola in a minimum 3-year rotation.

ABSTRACT

Clubroot (*Plasmodiophora brassicae*) on canola (*Brassica napus*) is spreading faster than expected in North Dakota, causing significant economic losses. An integrated management approach, including longer crop rotations, sanitation and cultivar resistance, are recommended to minimize the impact of this disease. Currently, cultivar resistance is the main management tool sought by growers for clubroot management, without longer rotations out of host crops. Short rotations with clubroot resistant (CR) canola in clubroot-infested regions may lead to resistance breakdown, eventually leading to a decline in canola hectares. The development of new CR cultivars, preferably carrying resistance to novel pathotypes of *P. brassicae*, is therefore important. To obtain cultivars resistant to the prevalent pathotypes, knowledge of pathotype distribution is necessary. Clubbed canola roots were collected from 32 infested fields in North Dakota, and representative samples were tested for pathotype designation on the hosts of the Canadian Clubroot Differential set. The *P. brassicae* pathotype composition in North Dakota was quite distinct from that reported previously from Alberta, Canada, where the clubroot outbreak is most severe. None of the pathotypes identified could overcome first generation resistance, and in North Dakota, clubroot may still be managed by planting CR canola in a minimum 3-year rotation.

INTRODUCTION

- *Plasmodiophora brassicae* causes clubroot on canola and is an emerging disease in North Dakota
- Can cause significant yield losses under favorable conditions (low pH soils, susceptible cultivar)
- Cultivar resistance, crop rotation and equipment sanitation are some of the common recommended practices to manage clubroot
- Planting resistant cultivars at shorter intervals increases the chances of resistance breakdown and development of novel pathotypes of *P. brassicae*
- Knowledge on the prevalent pathotypes in an area helps breeders to develop resistant cultivars and to develop integrated disease management guidelines
- The objective of this research is to determine the prevalent pathotypes of *P. brassicae* in North Dakota



Figure 1: Galls on canola roots due to *P. brassicae* infections in field were collected for pathotyping study

The host range of *Plasmodiophora brassicae* in North Dakota

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Abstract

Plasmodiophora brassicae causes clubroot on brassica crops and is a new emerging disease on rapeseed in North Dakota. A two-year study was conducted to document the host range and symptomatology on various brassica hosts to *P. brassicae* infections in field conditions. The results indicated that out of the 13 brassica hosts tested, 12 of them developed ellipsoidal galls on roots exhibiting the clubroot symptomatology with a disease index (DI) ranging from 41 to 100%. False flax/camelina (*Camelina sativa*) showed the least susceptibility among the brassica hosts tested. Symptomatology of clubroot on various brassica hosts will serve as a pictorial guide in the future to educate growers and in choosing non-brassica cover crops in clubroot infected fields.

Prevalent Pathotypes of *Plasmodiophora brassicae* in North Dakota

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¹Langdon Research Extension Center, North Dakota State University, Langdon, ND, 58249 U.S.A.,
²University of Alberta, Edmonton, AB, Alberta T6G 2P5, Canada.

MATERIALS & METHODS

- Clubbed galls from 32 canola fields were collected in annual survey of clubroot in North Dakota, USA
- Pathotyping was done under greenhouse conditions
- Six representative samples were evaluated for pathotype designation on the Canadian Clubroot Differential (CCD) set
- Thirteen differentials were inoculated with resting spores of *P. brassicae* and the experiment was repeated
- Galls on the differentials were evaluated after 45 days with clubroot

Table 1. Pathotype designations on the Canadian Clubroot Differential (CCD) set												
Differential	1	2	3	4	5	6	7	8	9	10	11	12
Pathotype	1	2	3	4	5	6	7	8	9	10	11	12
1	+	+	+	+	+	+	+	+	+	+	+	+
2	+	+	+	+	+	+	+	+	+	+	+	+
3	+	+	+	+	+	+	+	+	+	+	+	+
4	+	+	+	+	+	+	+	+	+	+	+	+
5	+	+	+	+	+	+	+	+	+	+	+	+
6	+	+	+	+	+	+	+	+	+	+	+	+
7	+	+	+	+	+	+	+	+	+	+	+	+
8	+	+	+	+	+	+	+	+	+	+	+	+
9	+	+	+	+	+	+	+	+	+	+	+	+
10	+	+	+	+	+	+	+	+	+	+	+	+
11	+	+	+	+	+	+	+	+	+	+	+	+
12	+	+	+	+	+	+	+	+	+	+	+	+

* Pathotype designations on the CCD set include a number to indicate the classification according to the system of Williams (1961), followed by a letter denoting the CCD designation (e.g., pathotype 2A, 2B, 2C) (Strelkov et al. 2020; Asanuma et al. 2020)
 * Since the CCD set includes all of the differentials of Somé et al. (1996), designations according to that system can also be obtained

RESULTS

North Dakota Clubroot Pathotype Designation				DISCUSSION	
Sample	Somé et al. (1996)	Williams (1961)	Canadian Clubroot Differential Set	<p>* The <i>P. brassicae</i> pathotype composition in North Dakota was quite distinct from that reported previously from Alberta, Canada, where the clubroot outbreak is most severe.</p> <p>* None of the pathotypes identified could overcome first generation resistance, and</p> <p>* In North Dakota, clubroot may still be managed by planting CR canola in a minimum 3-year rotation.</p>	
PCR	PS	8	Novel		
BBR	PS	2	2C		
BBR-2	PS	8	BN		
BBR-4	PS	8	Novel		
BBR-5	PS	8	ID	Acknowledgments	
BBR-6	PS	8	Novel	<p>We thank technical and review assistance of Dr. Strelkov lab personnel at University of Alberta, Edmonton, Canada and Special thanks to the support given by all the funding agencies: Northern Canola Growers Association, State Board of Agriculture Research and Education, ND Crop Protection Product Harmonization Board, and the Northern Canola Research Program (NIFA/USDA).</p>	

Literature Cited
 Asanuma H., Althausen A., Manoli V. P., Cao, Hwang S.-F., and Strelkov S. E. (2020). Vertical spread of single-spore and field isolates of *Plasmodiophora brassicae* able to overcome resistance in canola (*Brassica napus*). June 20, Plant Disease.
 Strelkov S. E., Hwang S. F., Manoli V. P., Turnbull G., Fredus-Ageman R., Kishna Hultman K. and Kaus S. (2020). Characterization of clubroot (*Plasmodiophora brassicae*) from canola (*Brassica napus*) in the Peace Country of Alberta, Canada. Canadian Journal of Plant Pathology. DOI: 10.1007/s40219-020-1778-1

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Summary

- Visual surveys indicate 20% of fields surveyed has Clubroot on Canola in Cavalier County in 2020
- Molecular studies of soil samples indicate 53% of the fields surveyed has clubroot resting spores in ND
- Clubroot Resistant Varieties are still holding good against the pathotypes present in ND soils
- Tested germplasm results are not presented
- Tested seed treatments had no efficacy in clubroot control
- Surfactants had an effect on Clubroot, however more testing has to be done
- Beet lime showed efficacy in all the rates tested
- Pathotyping studies are still being continued with University of Alberta, Edmonton, Canada

Acknowledgements

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- Mr. Barry Coleman and all the Canola Board members for their constant updates and guidance
- Dr. Strelkov, University of Alberta, Edmonton, Canada
- To the growers and collaborators across the state
- NDSU Soil testing lab



Thank You and Questions?