The Changing Face of Clubroot in Canada

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Outline of Presentation

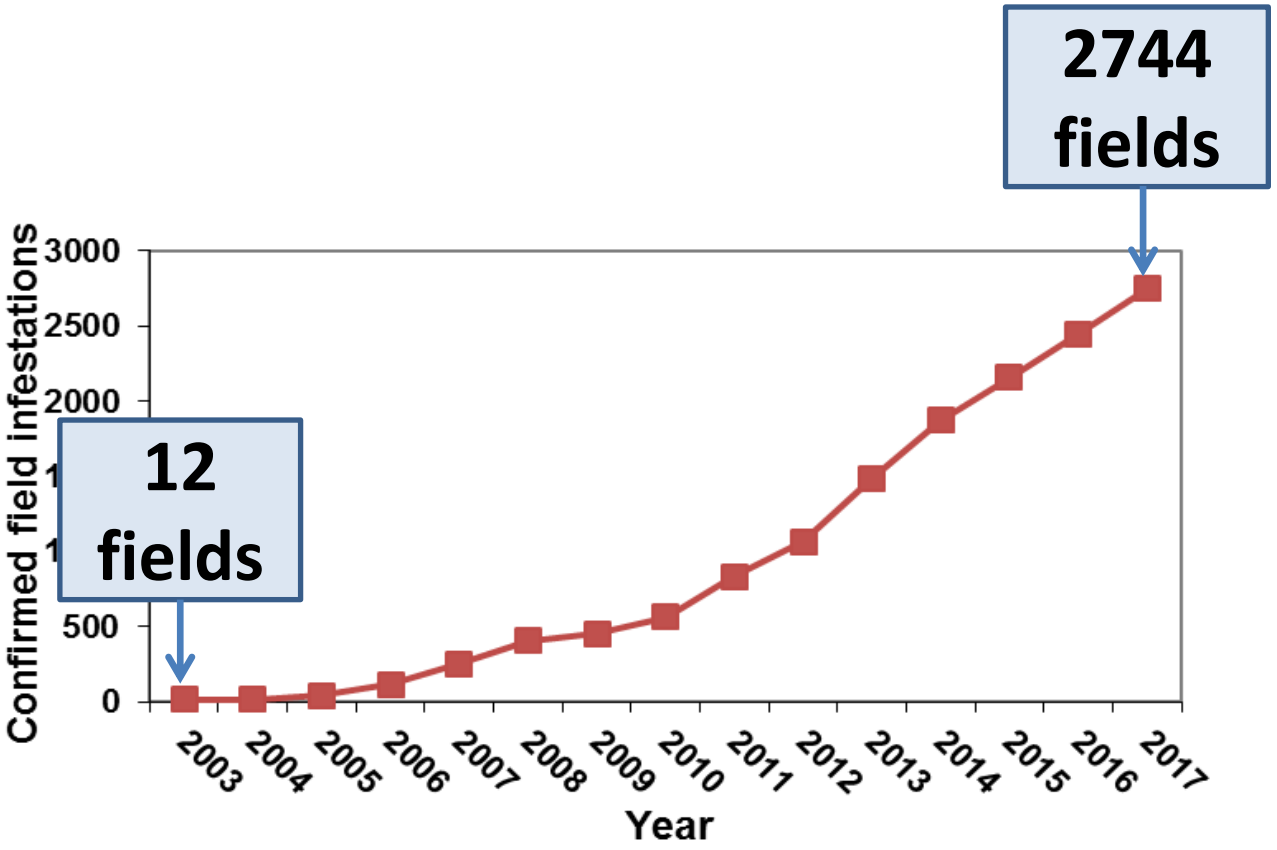
- Introduction
- Genetic resistance and “new” pathogen strains
- Pathotype identification
- Genetic diversity in *P. brassicae* populations
- Detection of new strains in old samples
- Conclusions

Introduction: Clubroot in Canada

- Likely introduced with infected fodder turnips
  - Well established by early 20th century
  - Constraint to cruciferous vegetable production in some regions
- Not identified on canola in western Canada until 2003

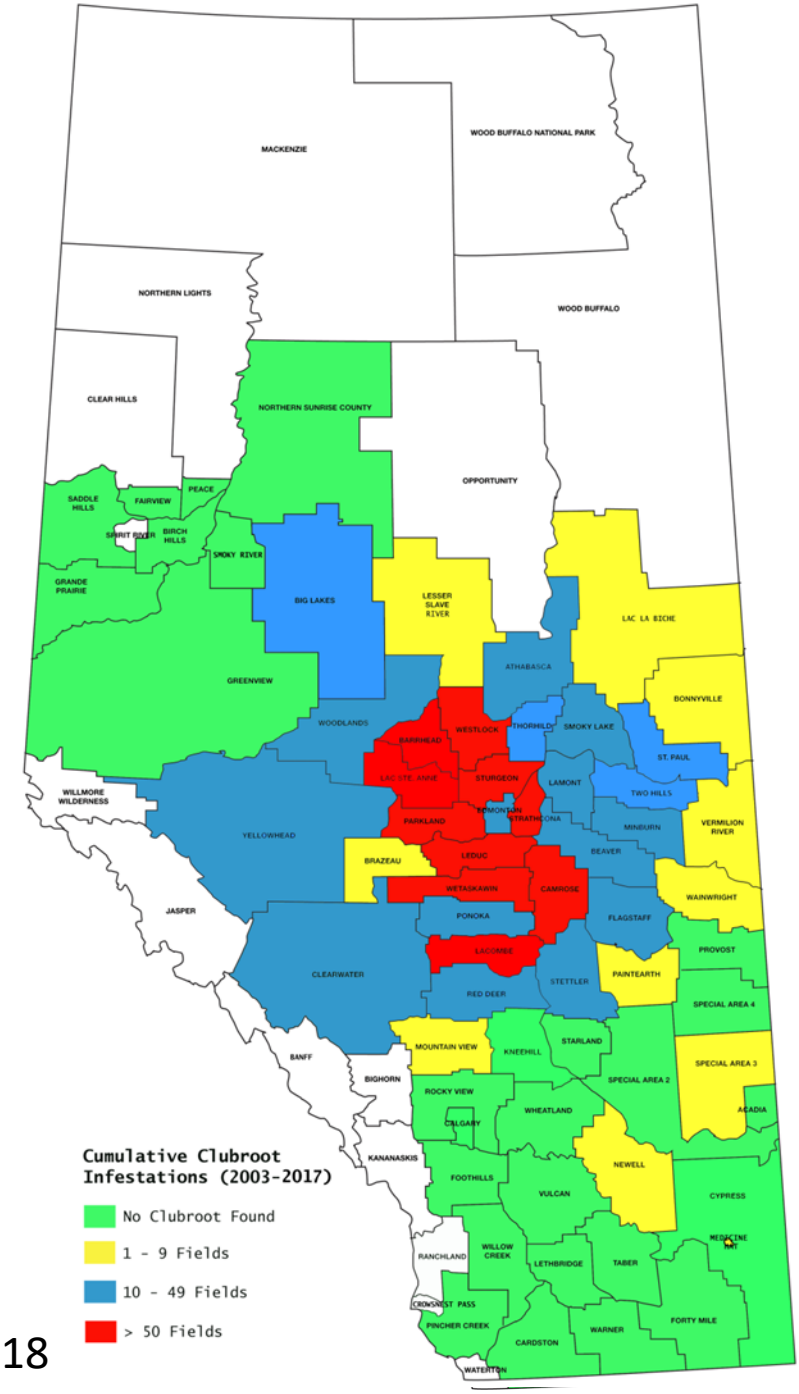
Clubroot Spread

- Spread has been rapid for a soilborne disease



Current Distribution

- Clubroot now found throughout much of Alberta
- Also increasing in Saskatchewan & Manitoba



Strelkov et al. 2018

Clubroot Management

- Relies mainly on planting clubroot resistant (CR) canola
- “Easiest” and most effective management method
  - Excellent control of all known pathotypes

New Strains of *P. brassicae*

- First CR canola variety released in 2009
- In 2013, patches of severe clubroot found in some fields planted with CR canola
- Testing confirmed presence of “new” strains that overcame resistance (Strelkov et al. 2016)

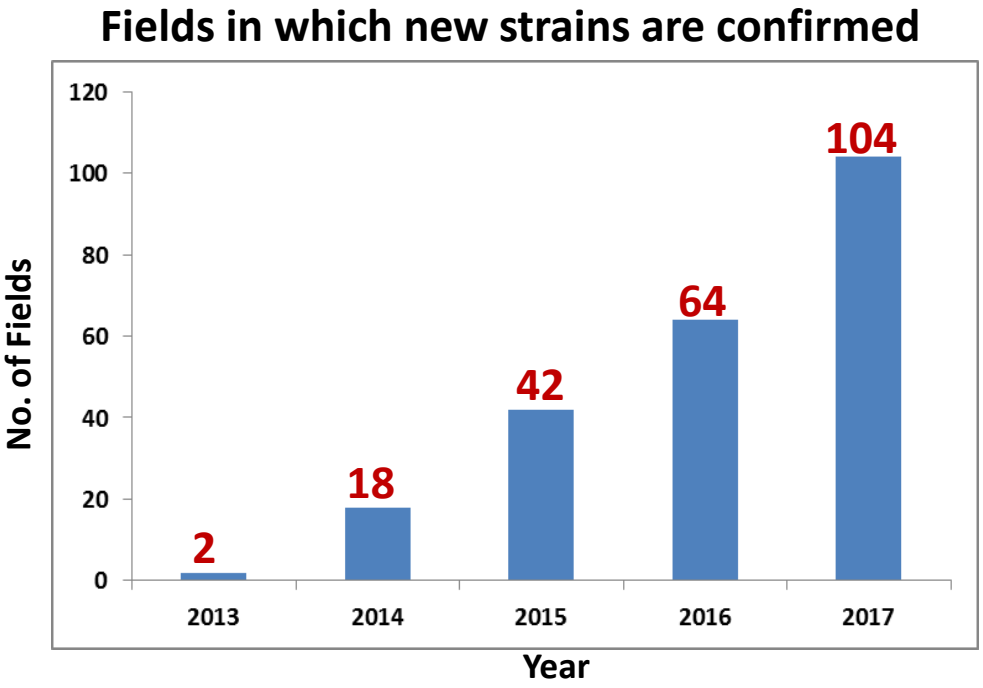
Photo: M. Harding

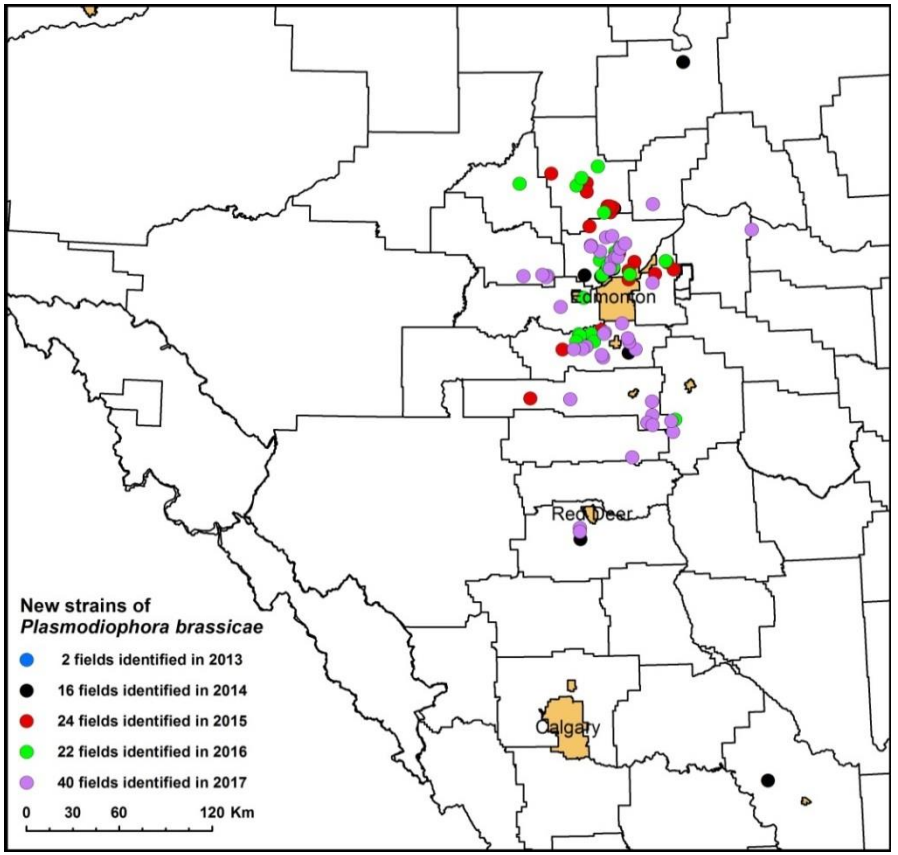
Clubroot in CR Canola

- Annual surveys have found increasing numbers of fields where resistance has been overcome
- Samples from each potential case are evaluated in a greenhouse in a 2-step process
  - Test against suite of CR canola varieties
  - If increased virulence detected, evaluate for pathotype designation

Clubroot Resistance Erosion

Increase in fields with resistance issues





Strelkov et al. unpublished

Samples tested from SK & MB do not appear to overcome resistance

Challenge: Pathotype Identification

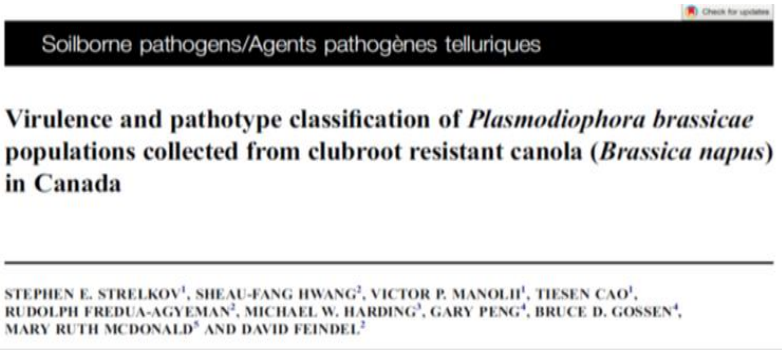
- "New" *P. brassicae* strains that overcome resistance cannot be distinguished from "old" strains based on commonly used pathotype classification systems
- Example: First of the new strains were classified as pathotype 5 on Williams' differential set
  - But this classification did not reflect their virulence on CR canola

Potential Confusion

- First of the new *P. brassicae* strains was referred to as pathotype '5X' to distinguish it from the old pathotype 5
- Soon, many people working with canola referred to all new strains as '5X'
  - Even after it became clear that not all new strains were alike & some had distinct virulence patterns!

Canadian Clubroot Differential Set

- Urgent need for a system to identify and distinguish *P. brassicae* strains
  - Keep up with the emerging new virulence phenotypes that were being identified
- Resulted in development of the Canadian Clubroot Differential (CCD) Set
  - Consists of 13 differential hosts: Williams, Somé et al., selected hosts of Europ
    Differential + several canol


Soilborne pathogens/Agents pathogènes telluriques

Virulence and pathotype classification of *Plasmodiophora brassicae* populations collected from clubroot resistant canola (*Brassica napus*) in Canada

STEPHEN E. STRELKOV[1], SHEAU-FANG HWANG[2], VICTOR P. MANOLII[1], TIESEN CAO[1], RUDOLPH FREDUA-AGYEMAN[2], MICHAEL W. HARDING[3], GARY PENG[4], BRUCE D. GOSSEN[4], MARY RUTH MCDONALD[5] AND DAVID FEINDEL[2]

CCD Pathotype Classifications

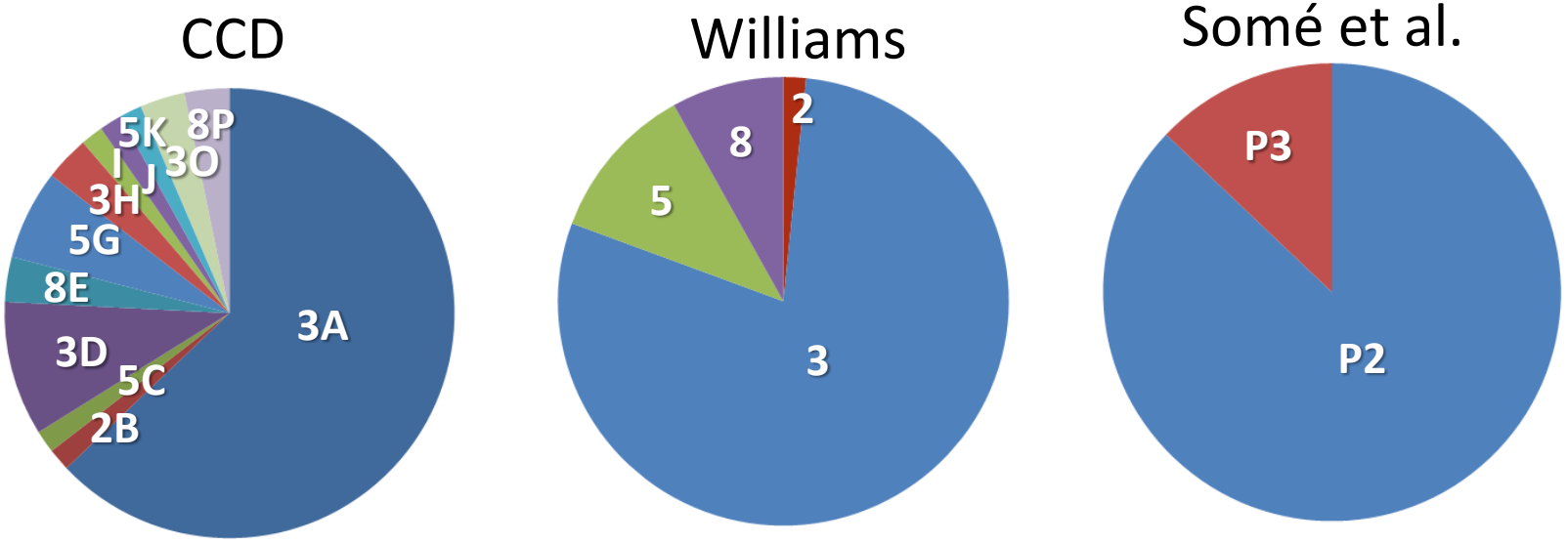
Differential Host	Reaction																
ECD 02	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
ECD 05	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ECD 06	+	+	+	+	+	+	-	+	+	-	-	-	+	+	-	+	-
ECD 08	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+
ECD 09	+	+	+	+	+	+	-	+	+	-	-	-	+	+	+	+	-
ECD 10 W	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
ECD 11 BS	-	+	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
ECD 13 JQ	+	+	-	+	-	+	-	+	-	-	-	-	+	-	+	-	-
Brutor	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Laurentian	+	+	-	+	+	+	-	+	-	+	-	-	-	+	+	+	-
Mendel	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+
Westar	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
45H29	+	+	+	+	+	-	+	-	-	+	+	-	-	-	+	+	+
Pathotype designations																	
CCD	A								I	J	K	L	M	N	O	P	X
Williams	3								5	8	5	5	6	8	3	8	5
Somé et al.	P2	P2	P2	P2	P2	P2	P3	P2	P2	P3	P3	P3	P2	P2	P3	P2	P3

CCD = pathotype A
Williams = pathotype 3
→ 3A

- Unique virulence patterns assigned different letters to designate each pathotype (Strelkov et al. 2018)
- Also allows for pathotype designations to be obtained as per Williams (1966) & Somé et al. (1996)

Pathotypes Identified 2014-2016

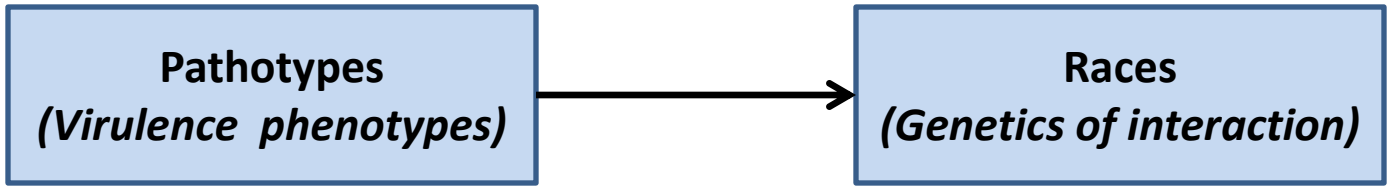
- CCD Set has a good differentiating capacity
- Enabled identification of multiple distinct virulence phenotypes among pathogen populations able to overcome resistance



Based on Strelkov et al. (2018)

Next Steps

- CCD Set can serve as the basis for a more refined classification system
  - **Short term:** removal of redundant differentials, replacement of one hybrid differential with non-hybrid carrying same resistance
  - **Longer term:** use of hosts with better defined resistance, near-isogenic lines as differentials



Genetic Diversity within *P. brassicae*

- Also interested in understanding the extent of genetic diversity within *P. brassicae* and relationships between pathogen populations
  - Used restriction site-associated DNA sequencing (RADseq) to examine diversity within *P. brassicae* single-spore and field isolates collected from across Canada (Holtz et al. 2018)


Holtz *et al. BMC Genomics* (2018) 19:254
https://doi.org/10.1186/s12864-018-4658-1

BMC Genomics

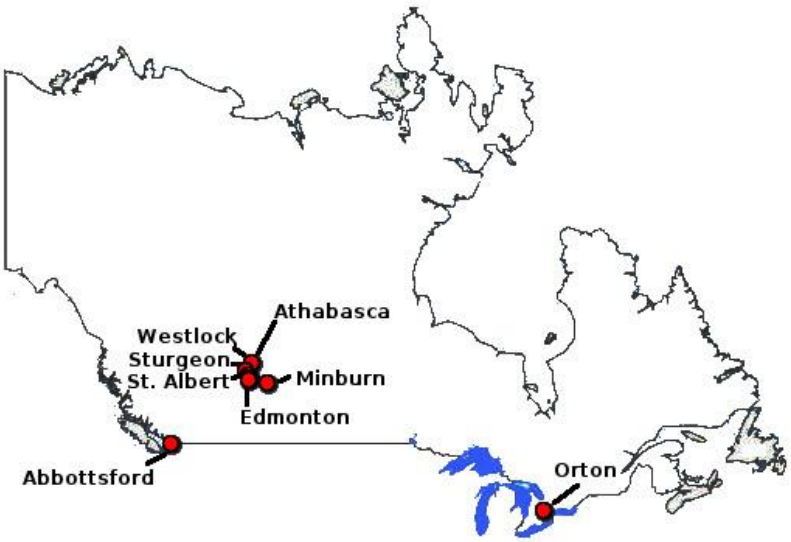
RESEARCH ARTICLE Open Access

Genotyping of *Plasmodiophora brassicae* reveals the presence of distinct populations

Michael D. Holtz[1], Sheau-Fang Hwang[2] and Stephen E. Strelkov[3*]

Isolates Studied

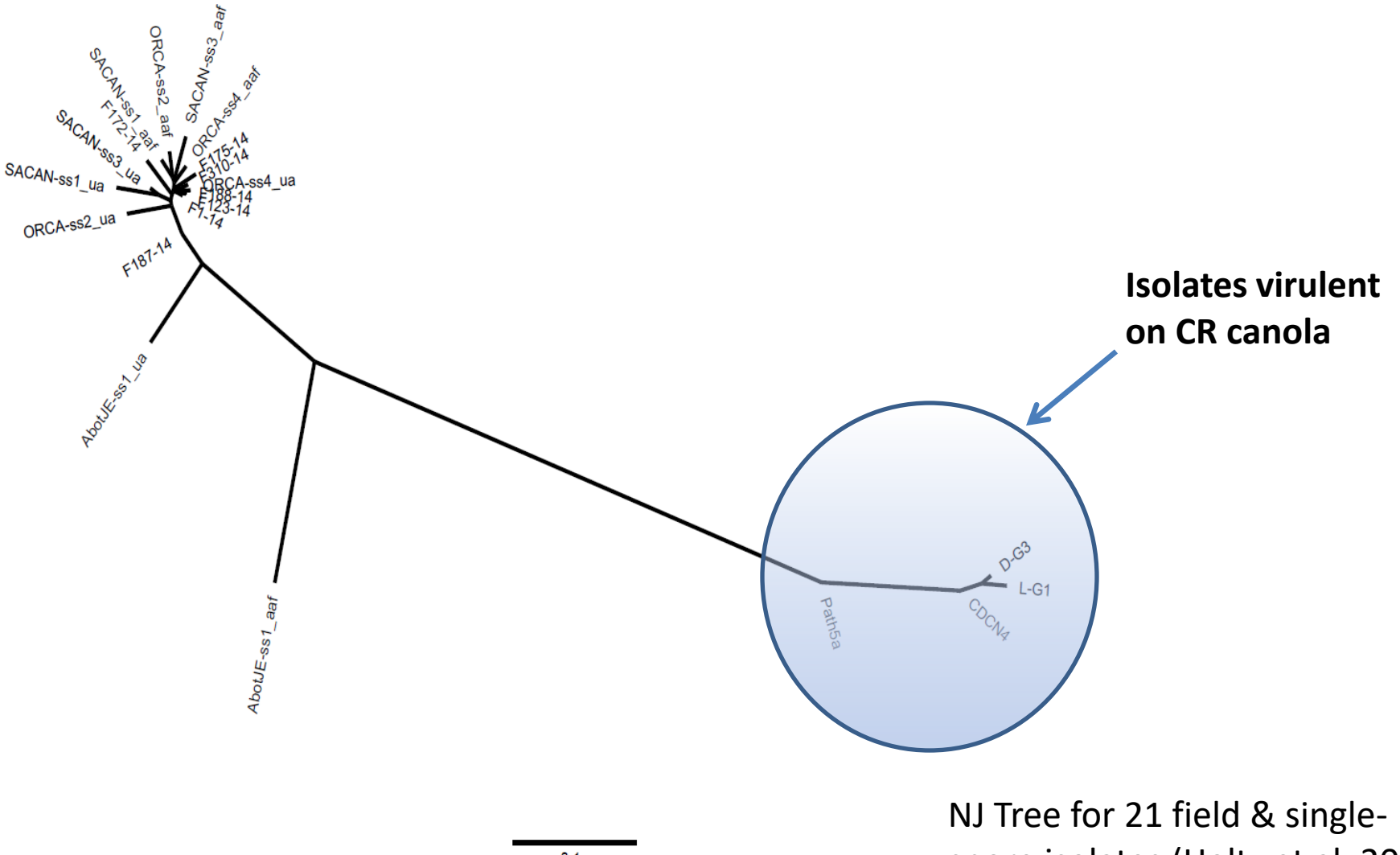
- Compared 21 field and single-spore isolates of *P. brassicae* from various geographical origins in Canada
- Included field isolates representing pathotype 5X



Results

- Population analysis indicated that most isolates belonged to one of two distinct populations
- Corresponded with the ability of isolates to cause disease on CR canola

Relationship Between Isolates



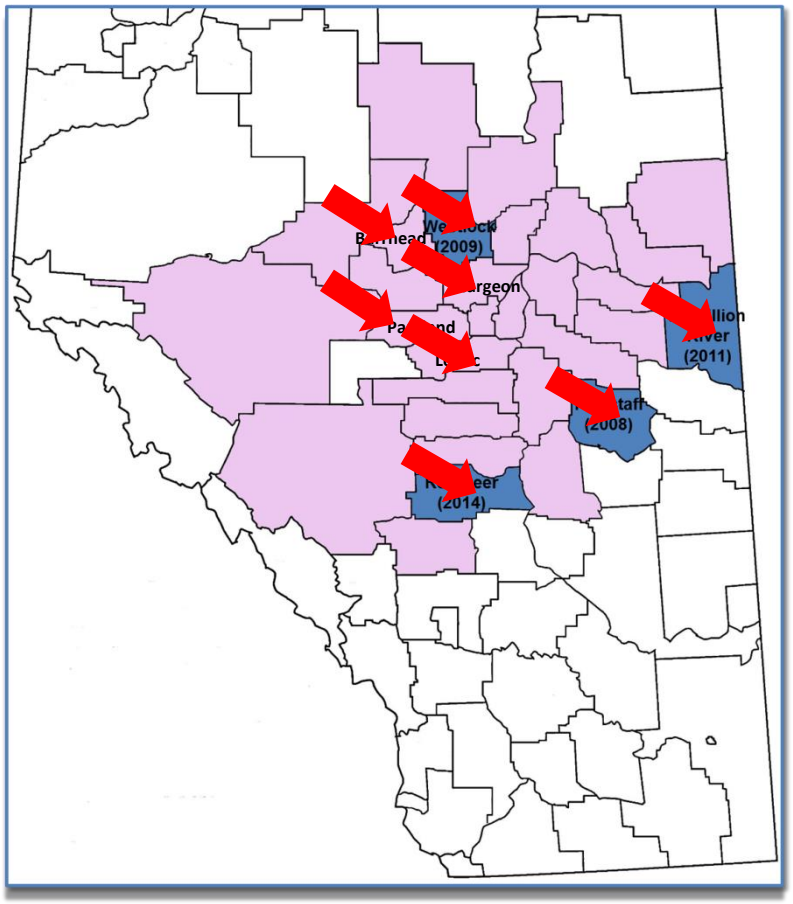
NJ Tree for 21 field & single-spore isolates (Holtz et al. 2018)

Are the “New” Strains Really New?

- The divergence between virulent and avirulent isolates has facilitated development of molecular markers to distinguish the pathogen populations
- Used these markers to look for the occurrence and distribution of the virulent population over time by examining root galls in our collection (2005-2016)

Occurrence of the New Strains

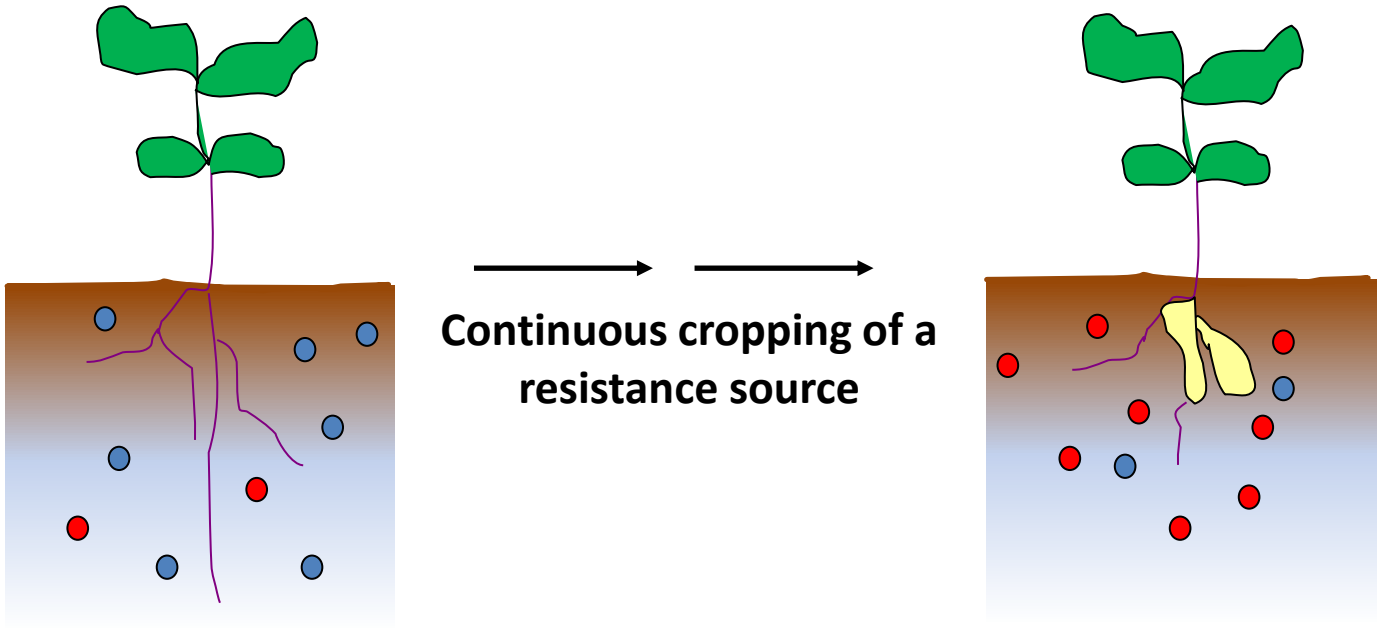
- Members of pathogen population virulent on CR canola widespread prior to its introduction
- Usually found at very low levels in galls from non CR-canola



Holtz et al. In preparation

Selection for New Strains

- Findings confirm hypothesis that proliferation of virulent strains resulted from selection pressure imposed by planting CR canola
  - Eventually resulted in pathotype shifts



Conclusions

- Canadian canola continues to be at risk from clubroot
- Genetic resistance is highly effective but also vulnerable to pathotype shifts
- Host differential sets and population genetics studies provide some insights into the pathogen
- Resistance stewardship and a more integrated approach will be needed for sustainable clubroot management

Acknowledgements

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