

Clubroot Resistance in Canola: Germplasm development and molecular mapping



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Overview

- **Search for resistance**
- **Germplasm development: Introgression of resistance**
 - Mendel resistance
 - Rutabaga resistance
 - Resistance from allied species
- **Molecular mapping of resistance**
 - Mendel resistance
 - Rutabaga resistance

Search for resistance

Hasan MJ, Strelkov SE, Howard MJ, Rahman H, 2012: Can J Plant Sci (in press)

Species	No. accession	No. of accession with resistance				
		Path 2	Path 3	Path 5	Path 6	Path 8
<i>B. rapa</i> var. <i>rapifera</i>	5	5	5	5	5	5
<i>B. rapa</i> var. <i>chinensis</i>	8	1	1	2	1	1
<i>B. rapa</i> var. <i>pekinensis</i>	5	0	0	0	0	0
<i>B. rapa</i> var. <i>oleifera</i>	18	6	9	9	12	12
<i>B. rapa</i> var. <i>oleifera</i>	9	0	0	0	3	3
<i>B. oleracea</i> var. <i>capitata</i>	16	1	3	2	1	2
<i>B. oleracea</i> var. <i>botrytis</i>	14	0	0	0	0	0
<i>B. oleracea</i> var. <i>italica</i>	13	0	0	0	0	1
<i>B. oleracea</i> var. <i>gemmifera</i>	3	0	2	1	0	0
<i>B. oleracea</i> var. <i>alboglabra</i>	2	0	0	0	0	0
<i>B. oleracea</i> var. <i>villosa</i>	1	0	0	1	0	0
<i>B. nigra</i> (different ssp.)	77	67	70	67	68	75
<i>B. napus</i>	36	29	3	1	27	32
<i>B. napus</i> ssp. <i>napobrassica</i>	5	5	3	4	5	5
<i>B. juncea</i>	48	0	0	0	0	0
<i>B. carinata</i>	24	0	0	0	0	0
Total	275					



Germplasm development: Mendel resistance

Rahman et al. 2011: Can J Plant Sci 91:447-458



Two crosses → 1,872 F₂ plants (sel. for CR)
→ 18+22 = 40 F₃ families

Cross	No. F ₃ fam.	No. segr. fam.	No. non-segr. fam.	χ ² (1:2 segr.)	p
Mendel x A04-71NA	18	8	10	1.00	0.3-0.5
Mendel x A04-75NA	22	16	6	15.52	<0.05

	Total F ₃ plants	% Res. plants	No. R-plants flowered	Days to flower	
				Range	Mean
Mendel x A04-71NA	296	83.4	77	47 - 70	63.4
Mendel x A04-75NA	313	95.5	37	49 - 64	56.4
<i>Hi-Q</i>				46 - 48	47.0



Germplasm development: Mendel resistance

Field evaluation of F₅ families in Leduc in 2008



	Clubroot res.		Days to flower		% Oil		Saturated FA	
	Range	Mean	Range	Mean	Range	Mean	Range	Mean
Mendel x A04-71NA	0.0-3.0	0.43	44-52	47.1	45.0-52.7	48.9	6.6-8.0	7.26
Mendel x A04-75NA	0.0-3.0	0.40	43-51	46.0	45.9-55.8	51.7	6.2-7.4	6.85
<i>46A65</i>	<i>3.0</i>	<i>3.00</i>	<i>44-45</i>	<i>44.5</i>	<i>50.8-53.2</i>	<i>52.5</i>	<i>6.8-7.0</i>	<i>6.90</i>

Index of disease: Resistant F₆ lines & hybrids – few examples

	Path 3	Path 5	Path 6
591.165 OP	0.0	0.0	0.0
591.165 Hybrid	0.0	0.0	7.4
591.186 OP	0.0	0.0	0.0
591.186 Hybrid	0.0	0.0	0.0
591.225 OP	27.8	0.0	0.0
591.225 Hybrid	22.2	0.0	9.3
591.232 OP	0.0	0.0	0.0
591.232 Hybrid	0.0	2.8	0.0

Some of these F₆ and F₈ lines supplied to interested canola Breeders



Germplasm development: Mendel resistance – 2nd cycle breeding

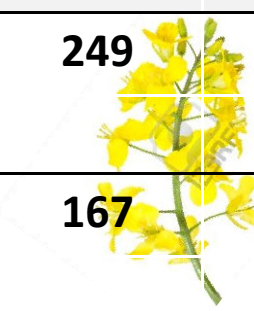
Clubroot resistant x Susceptible HT canola



Gene- ration	No. plants tested for HT & CR
F₂	1144
F₃	877

Evaluation F₅ families in field 2010

	No. F ₅ fam.	Days to flower	% Oil	% Protein	Glucosi- nolate	% Sat FA
RoundUp	249	45 - 50	44.4 - 53.5	19.7 - 25.8	11.2 - 19.5	5.8 - 7.7
<i>A07-26NR</i>		<i>45.2</i>	<i>50.6</i>	<i>22.1</i>	<i>13.9</i>	<i>6.5</i>
Clearfield	167	44 - 51	40.4 - 52.8	21.7 - 28.7	9.5 - 22.5	5.8 - 7.8
<i>A05-17NI</i>		<i>44.6</i>	<i>48.3</i>	<i>25.1</i>	<i>12.8</i>	<i>6.3</i>



Germplasm development: Mendel resistance – 2nd cycle breeding



2011 Field trial data of clubroot resistant F₆ breeding lines

[RoundUp (n=47), 2 loc; Clearfield (n=30), 3 loc; data of top 1/3rd – 2/3rd lines]

		Days to flower	Yield (%)	% Oil	% Protein	% Sat FA
RoundUp	Range	54.5 - 56.3	91.2 - 105.1	46.7 - 50.9	22.3 - 26.0	6.16 - 7.18
	Mean	55.5	96.8	48.6	23.9	6.74
A07-26NR (OP)	<i>Mean</i>	55.5	104.8	51.5	21.8	6.79
95-93 (hybrid)	<i>Mean</i>	55.8	95.2			
Clearfield	Range	53.0 - 56.2	91.3 - 107.9	45.8 - 50.5	23.9 - 26.4	6.16 - 7.17
	Mean	54.7	98.2	47.6	25.4	6.76
A05-17NI (OP)	<i>Mean</i>	54.0	96.2	46.9	25.9	6.64
45P70 (hybrid)	<i>Mean</i>	54.2	103.8			



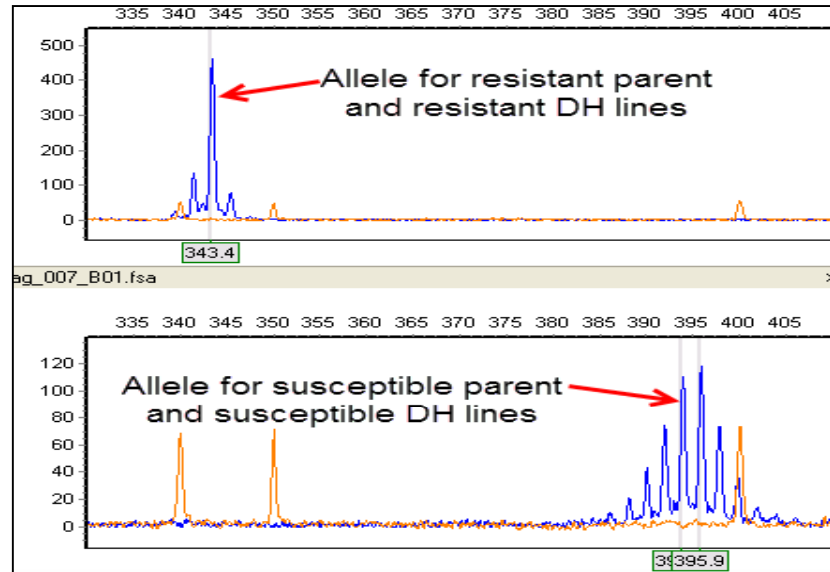
All RoundUp and Clearfield lines are canola quality

Mendel resistance: Molecular mapping

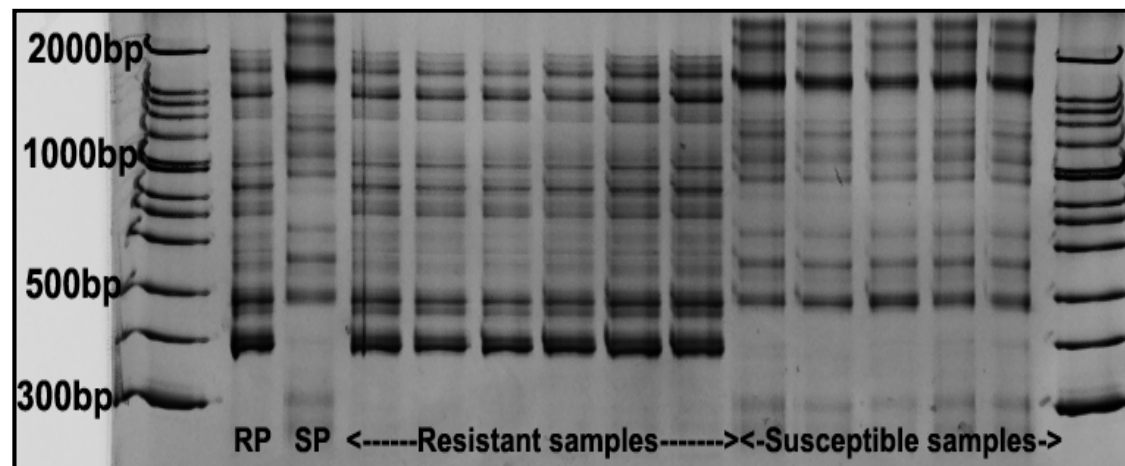
- Resistant x Susceptible crosses
- Four (2 DH + 2 pedigree) mapping populations developed
- SSR markers used



ABI genotyping electropherogram



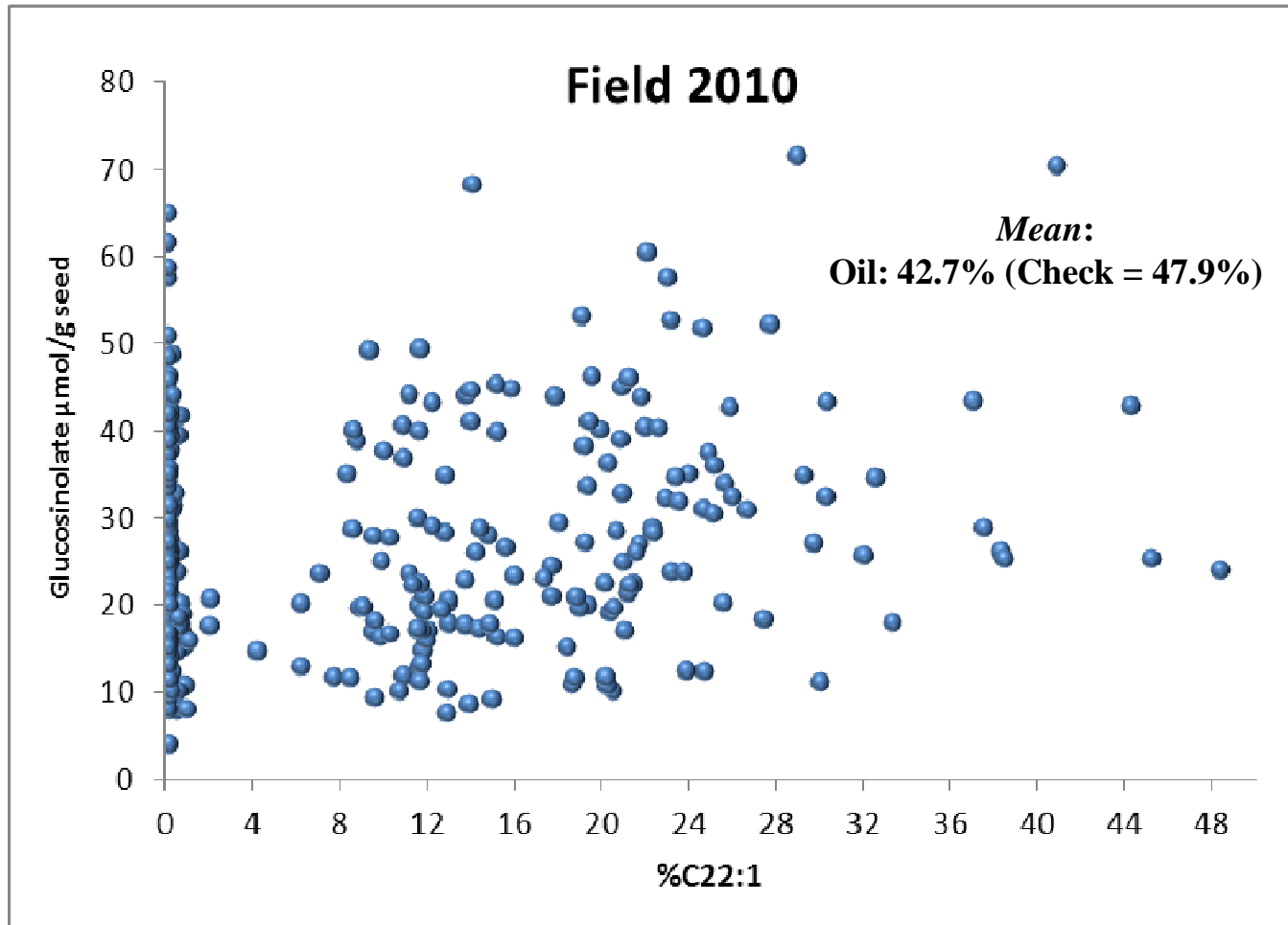
Polyacrylamide gel image



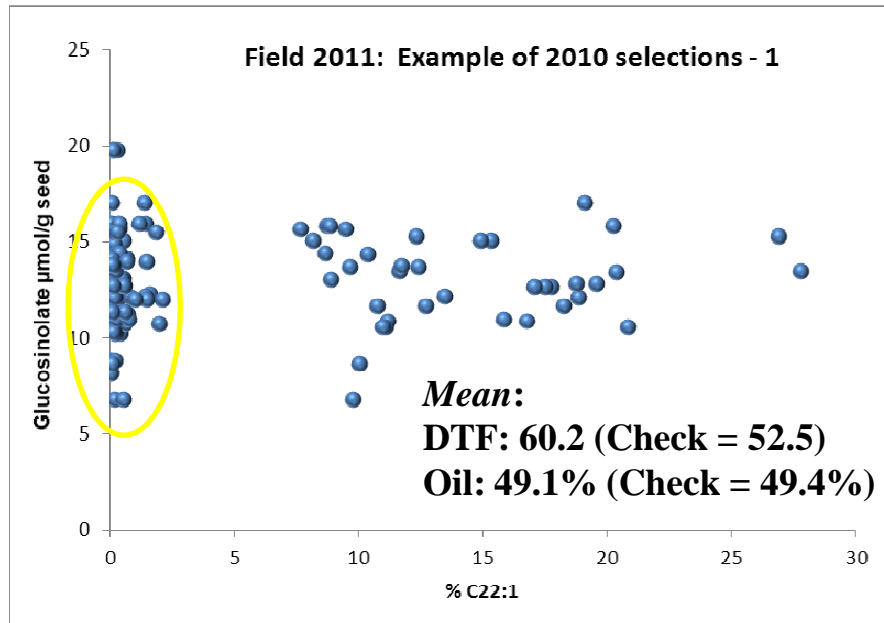
Germplasm development: Rutabaga resistance

Rutabaga x Canola crosses: *Conventional and HT populations*

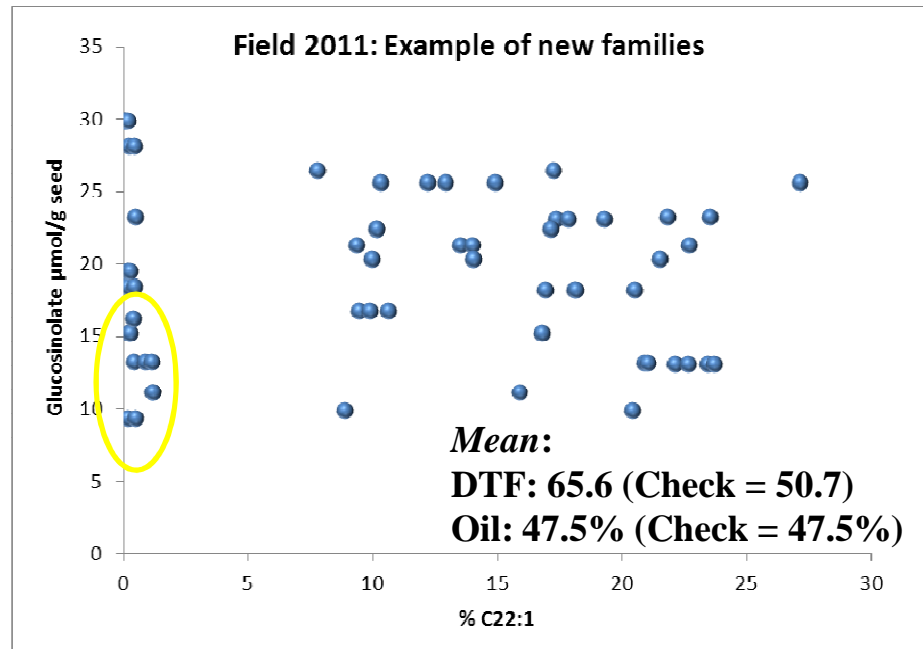
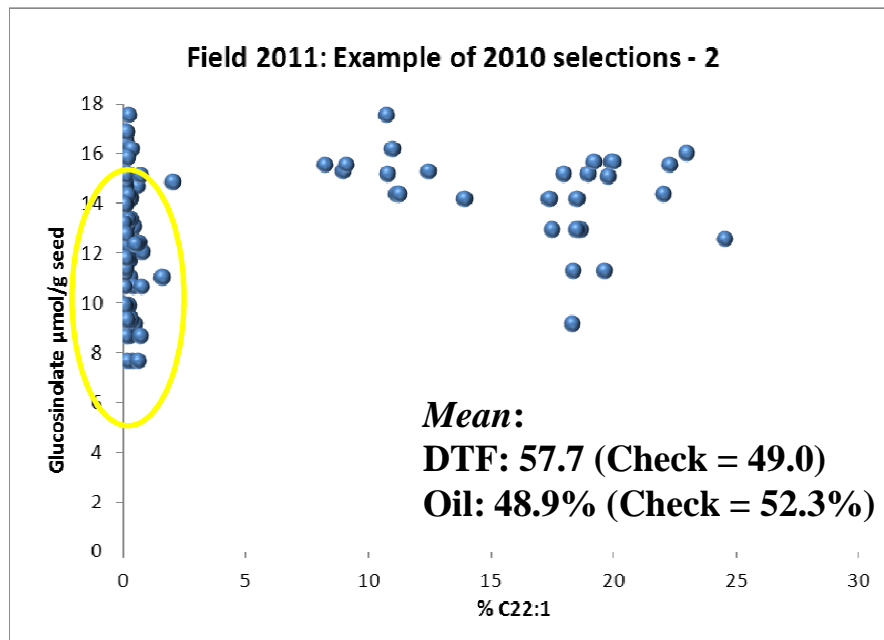
No. pedigree families ($\approx F_3$) in Field test in 2010 = 209



Germplasm development: Rutabaga resistance



**Number pedigree families ($\approx F_4$)
in Field test in 2011:
204 families from 2010 selection +
179 new families = 383**



Germplasm development: Rutabaga resistance

Rutabaga x Canola crosses: Conventional and HT Canola line development in greenhouse



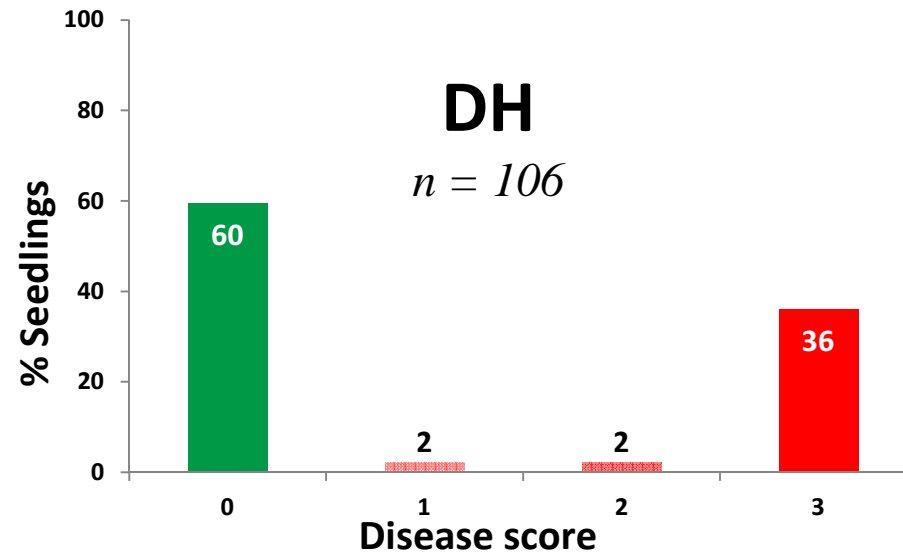
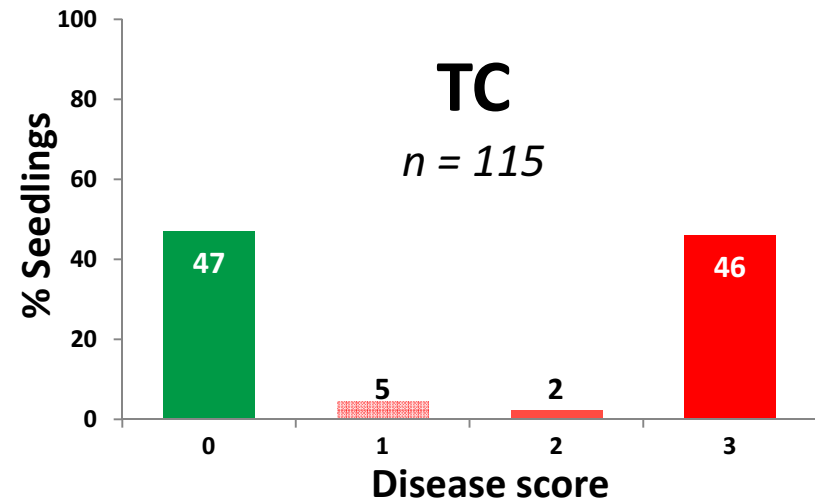
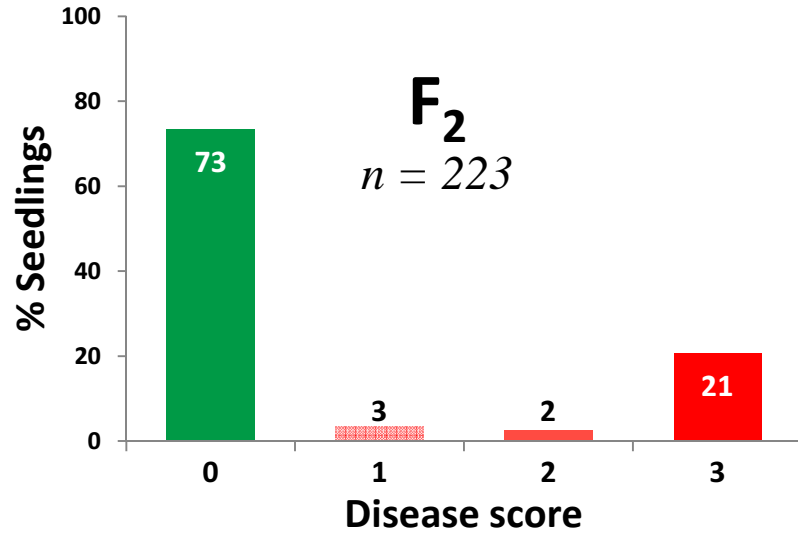
Popula- tion		No. plants	No. resistant	GLS	%C22:1	%Sat
#1 ¹	Range	3799	-	-	0.1 – 29.5	6.3 – 11.0
	Mean				11.8	7.3
#1-progeny ²	Range	440	-	7.8 – 82.2	0.03 – 22.8	6.0 – 9.9
	Mean			11.8	5.4	7.6
#2 ³	Range	1242	478	-	0.04 – 14.7	6.7 – 9.7
	Mean				5.6	8.1
#3 ⁴	Range	1049	137	-	0.04 – 16.0	6.4 – 10.2
	Mean			-	6.6	8.2
Total		6,530	615			

¹n = 304 for %C22:1 & %Sat; ²n = 106 for GLS, and 140 for %C22:1 & %Sat; ³n = 220 for %C22:1 & %Sat; ⁴n = 58 for %C22:1 & %Sat



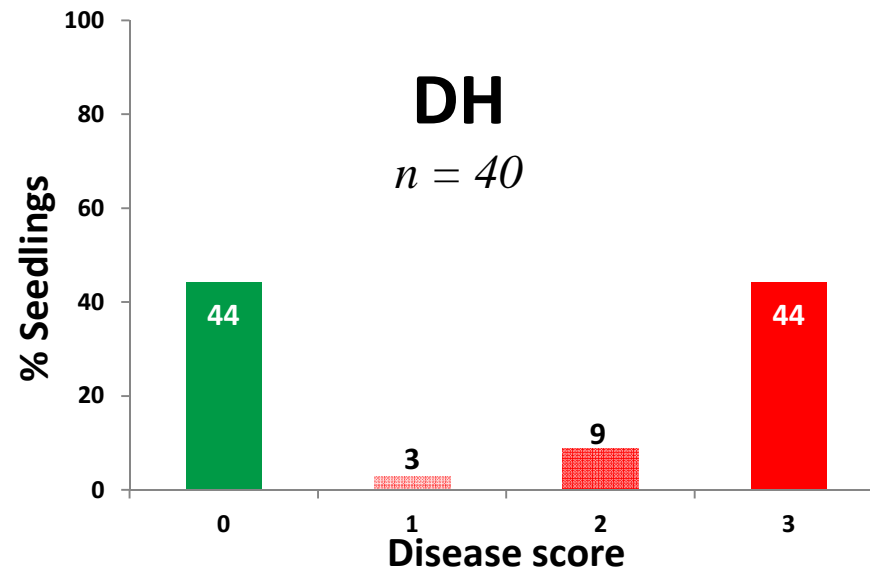
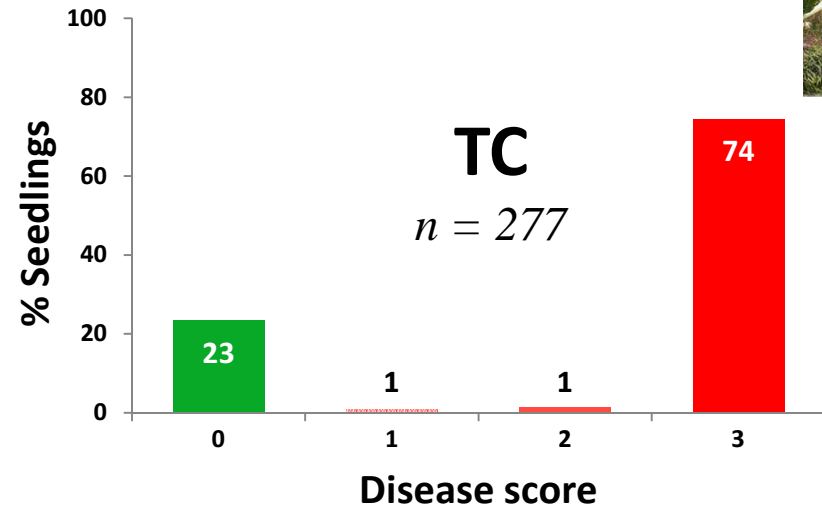
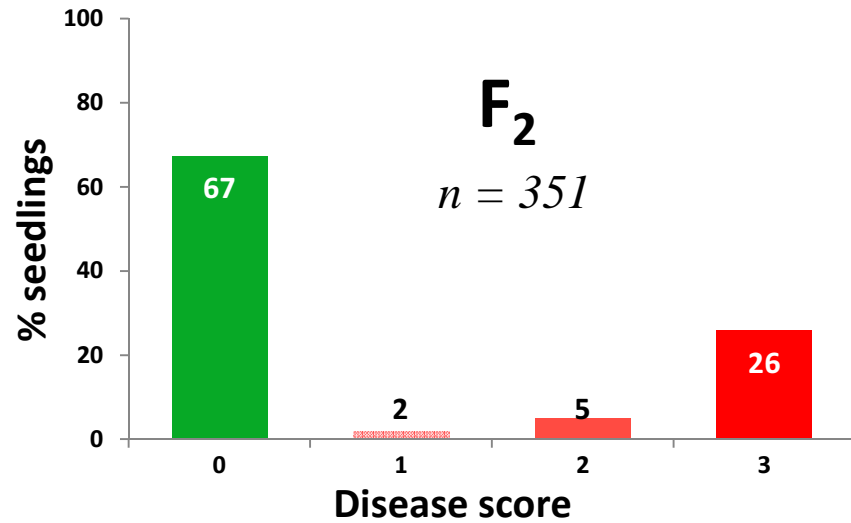
Rutabaga resistance: Molecular mapping

Resistance to pathotype 3: Rutabaga x A07-29NI



Rutabaga resistance: Molecular mapping

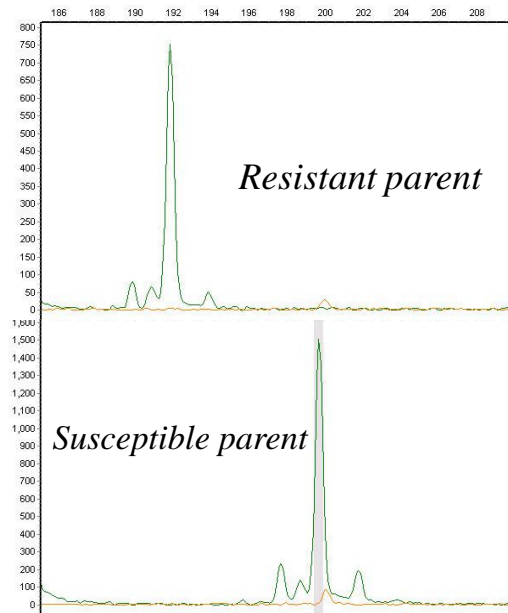
Resistance to pathotype 3: Rutabaga x A05-17NI



Rutabaga resistance: Molecular mapping



Rutabaga x Canola mapping populations *Polymorphism of SSR markers*



**ABI genotyping
electropherogram**

	Total SSR tested	% poly- morphic (> 4 bp)
Rutabaga-PL x A05-17NI	283	36.6
Rutabaga-BF x A07-29NI	271	35.1
Not amplified or < 4 bp	219	28.3
Total	773	

*Two genomic regions identified through
single marker analysis*



QTL mapping in progress

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