



New Sources of Resistance to Fusarium Head Blight and Indicators of Their Mode of Action

G. Fedak¹, W. Cao¹, D. Chi¹, D. Somers², A. Malhipour³, S. Miller¹, T. Ouellet¹, A. Xue¹, J. Gilbert³, M. Savard¹ and H. Voldeng¹

¹. Eastern Cereal and Oilseed Research Centre, AAFC, Ottawa, ON K1A 0C6
Canada

². Vineland Research and Innovation Centre, Vineland ON L0R 2E0 Canada

³. Cereal Research Centre AAFC, Winnipeg MB R3T 2M9 Canada



120 BC2F3 entries x 3 reps – Ottawa ON





Greenhouse misting



Cabinet misting system





Outline

Fusarium head blight resistance from wide crosses

- Hexaploid wheat
 - *Tritium monococcum*
 - *Aegilops speltoides*
 - *Triticum timopheevi*
 - *Triticum miguschovae*
 - *Aegilops cylindrica*
- Tetraploid wheat
 - *Tritordeum*
 - *Triticum carthlicum*
- Triticale

Thinopyrum elongatum

- GFP studies
- Microarray analysis

- Accession PI343447 crossed & backcrossed to Crocus.
- 535 plants advanced to F7 by SSD.
- TC67 selected.
- Mapping population of 230 F7 RIL lines produced from TC67 X Brio cross



Spikes of a *T. timopheevi* derivative at 21 days after point inoculation

FHB symptoms on TC67(*T. timopheevi* derivative) and check cultivars(2 year data)

Line	Incidence	Severity	FDK(%)	DON (ppm)
Sumai3	14.8	6.3	9.5	6.6
Hy644	42.6	20.0	35.1	25.3
Roblin	90.7	81.9	66.1	35.0
TC67	29.4	16.9	14.5	7.7

A QTL on 5A Chromosome in TC 67

Development of plant materials



TC 67 x **Brio**
($2n=6X=42$, AABBDD) ($2n=6X=42$, AABBDD)

F₁

SSD

F₇

230 lines



Field evaluation

- **To detect resistance to:**
 - initial infection (type I)
 - disease spread (type II)
 - DON accumulation (type III)
 - FDK (type IV)
- Spray inoculation was used
- Disease incidence, severity, index, DON accumulation, and FDK were measured



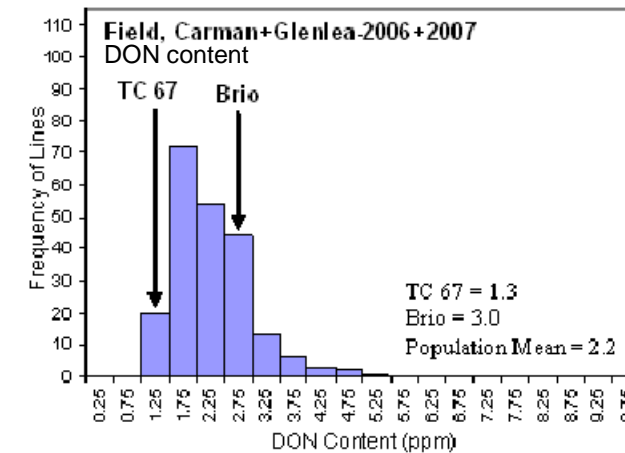
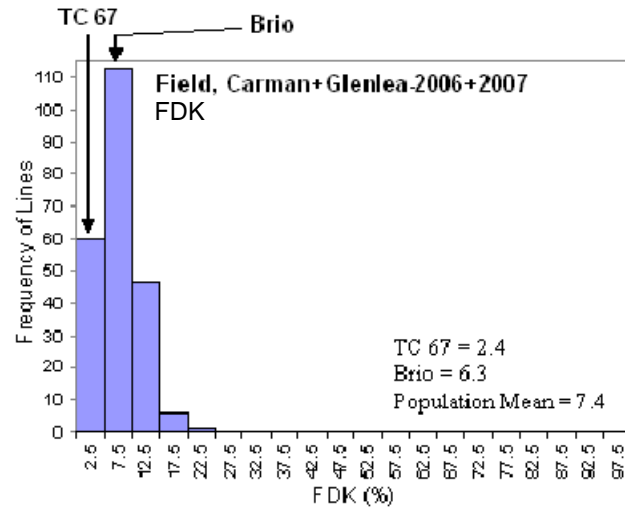
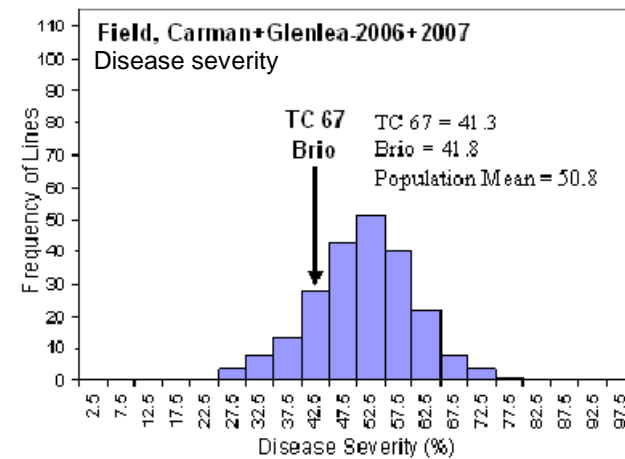
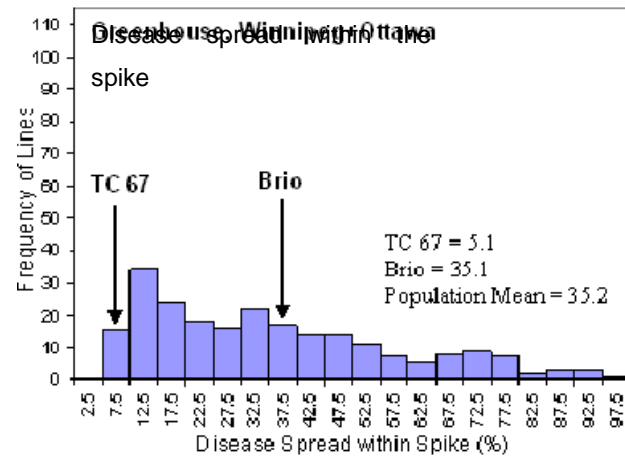
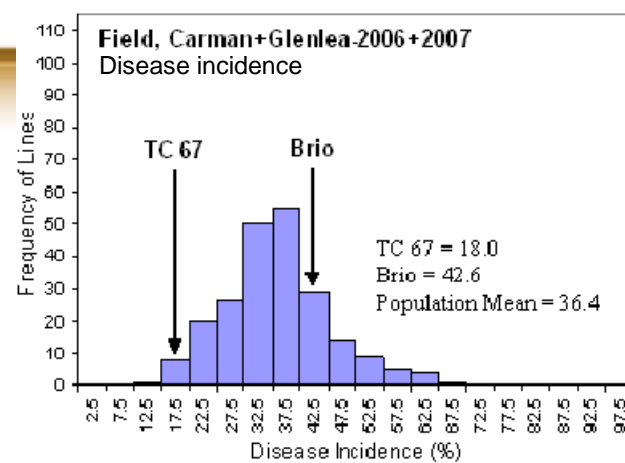
Greenhouse evaluation

- To detect resistance to:
 - disease spread (type II)
- Single-floret inoculation was used
- Disease spread was measured



Results and Discussion

Frequency distribution



Statistical analysis

- SAS®

Genotypic data collection

- DNA extraction
- DNA amplification: SSRs
- PCR products: ABI Analyzer

Bulked segregant analysis

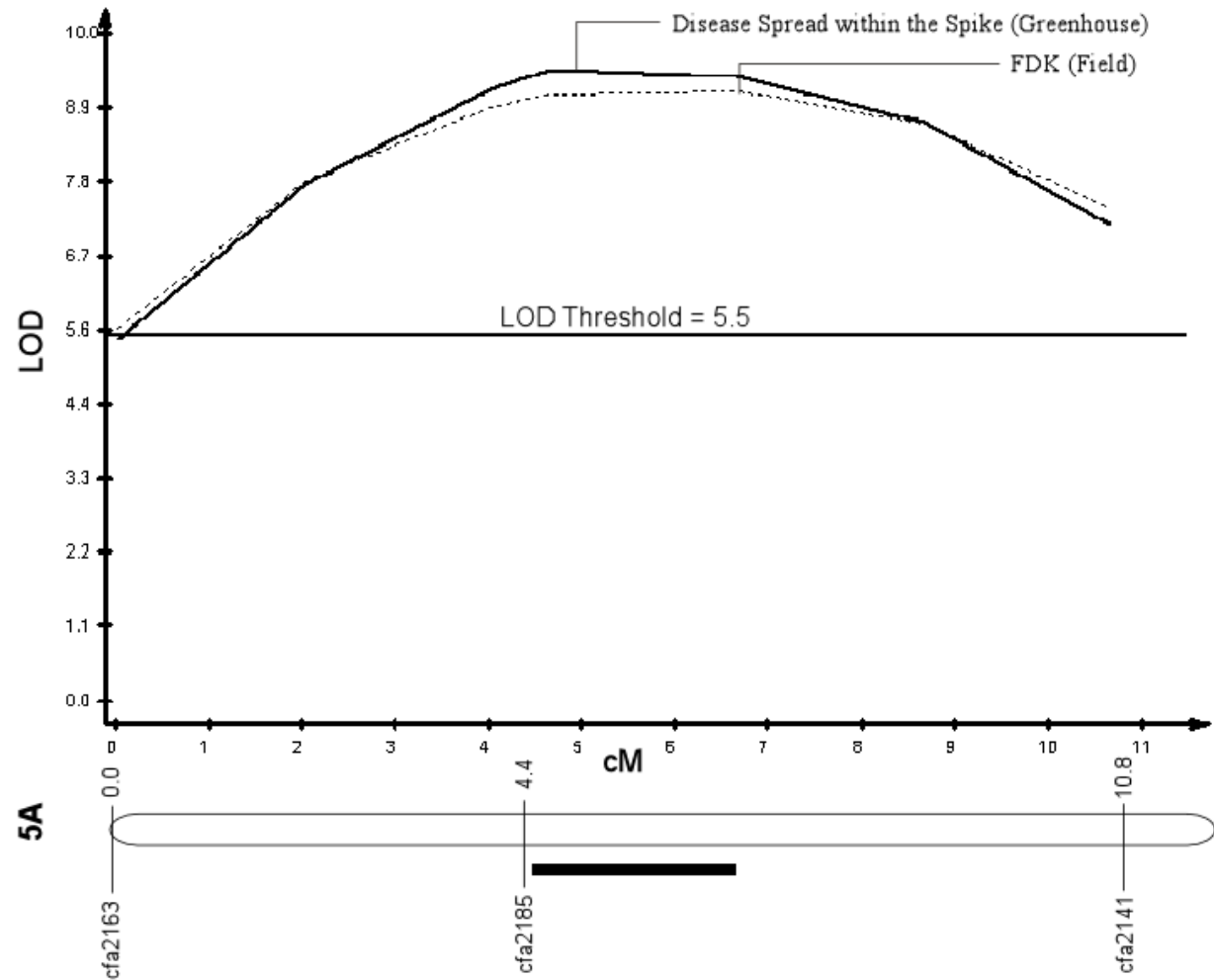
- 851 SSR primers
- Primers screened on:
RP, SP, RB, and SB
- Polymorphic markers screened
on: individuals of RB and SB
- Highly polymorphic markers
screened on: entire population

QTL mapping

- MAPMAKER/EXP
- QTL Cartographer

Results and Discussion

QTL mapping



- 200 accessions screened.
- Accession 10-1 crossed & backcrossed to AC Superb.
- DH mapping population produced from cross to AC Superb



T. monococcum

- 50 accessions
screened, 3
backcrosses to AC
Superb required to
restore fertility.

- DH mapping
population produced
from cross to AC
Superb.



Aegilops speltoides

Average metaphase I chromosome pairing in F1 hybrids between *Triticum aestivum* (c.v Superb) and *Aegilops speltoides* accession

Metaphase I configurations

I	II			III	IV	Xta freq.	No of cells
	Rod	Rings	Total				
9.55	4.82	2.40	7.22	1.29	0.14	11.90	88
10.80	4.60	3.10	7.70	0.36	0.18	12.62	112

FHB symptoms and DON content of progenies from interspecific crosses with bread wheat (AC Superb) (field data, 2005)

Source of resistance	Generation	Number of lines	FHB Index (range)	DON Level (range) ppm
<i>T. monococcum</i>	BC ₂ F ₆	22	0.3-3.0	0.1-1.7
<i>Ae. speltoides</i>	BC ₃ F ₆	70	0.3-7.5	0.3-2.0
Checks				
Roblin			76.5	9.1
Superb			24.0	9.2
Fukuho			1.5	1.9
Sumai3			1.3	2.9



M 321



S 184



AC Barrie



M 321

- Single accession crossed & backcrossed to AC Superb & progeny advanced to F6 by SSD.

- Resistant line selected.

- DH production in progress.



Triticum miguschovae

Average metaphase I chromosome pairing in F1 hybrids between *Triticum aestivum* (c.v.Superb) and *T. miguschovae* (AGD genome)

Metaphase I configurations

I	II		Total	III	IV	Xta freq.	No of cells
	Rod	Rings					
4.61	3.81	13.75	17.56	0.57	0.14	32.87	150

FHB symptoms and DON content in progenies from intercrosses of bread wheat with *T. miguschovae* (AGD) (field data 2005)

Generation	Number of lines	FHB Index (range)	DON Content (range) ppm
BC₃F₄	7	0.3-4.0	0.2-3.4
Checks			
Roblin	–	81.0	9.1
Superb		13.5	9.2
Fukuho		0.5	1.9

Symptoms on BC2 spike
of hybrid between Superb
and *T. miguschovae* at 21
days after point
inoculation



Fusarium Head Blight Resistant Lines Derived From Hybrids with *Aegilops cylindrica*

Hybrid combination	Generation	Number of lines
AC Superb X 1/74-91 X 5/20-91	F6	220
Teal	F2-F5	bulks
AC Barrie		
Domain		
Elsa		
H4644		
Glenlea		
Alsen		

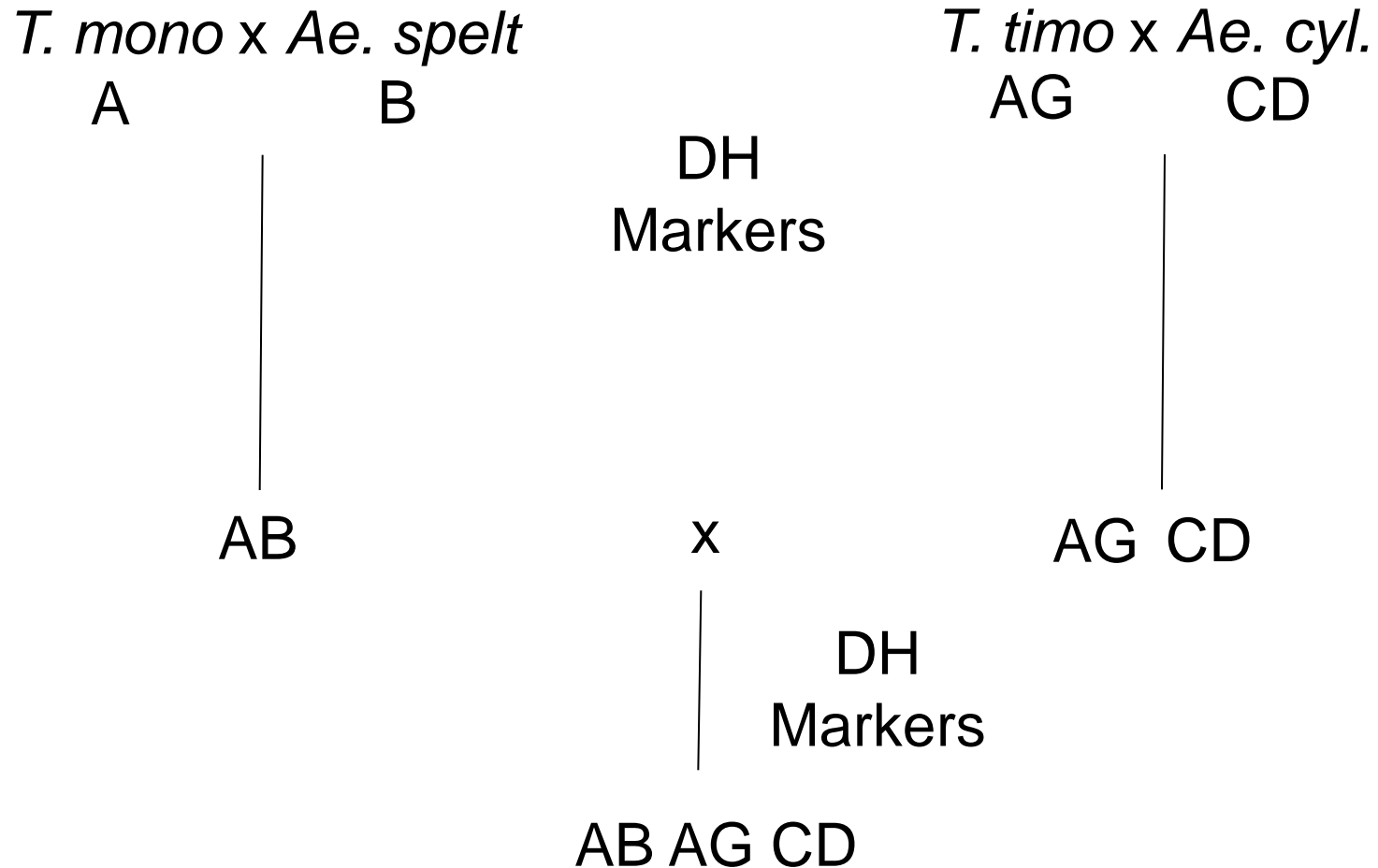
FHB Symptoms in Derivatives from *Triticum miguschovae* and *Aegilops cylindrica*

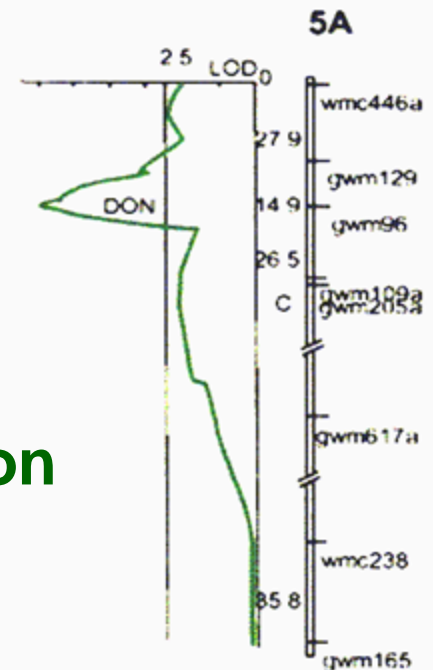
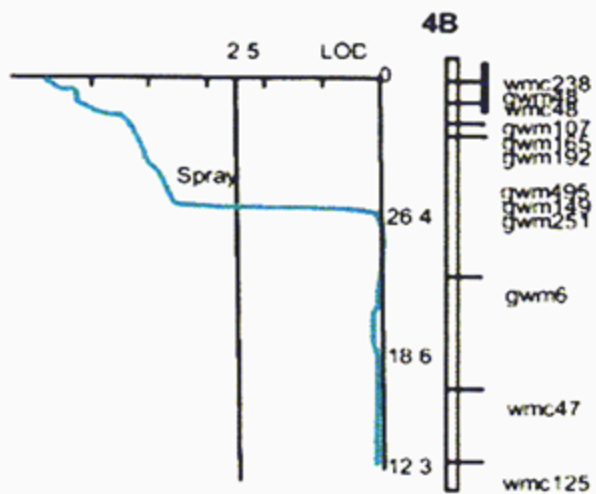
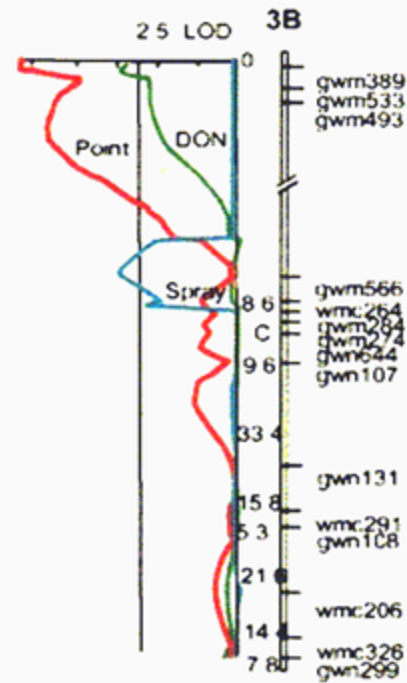
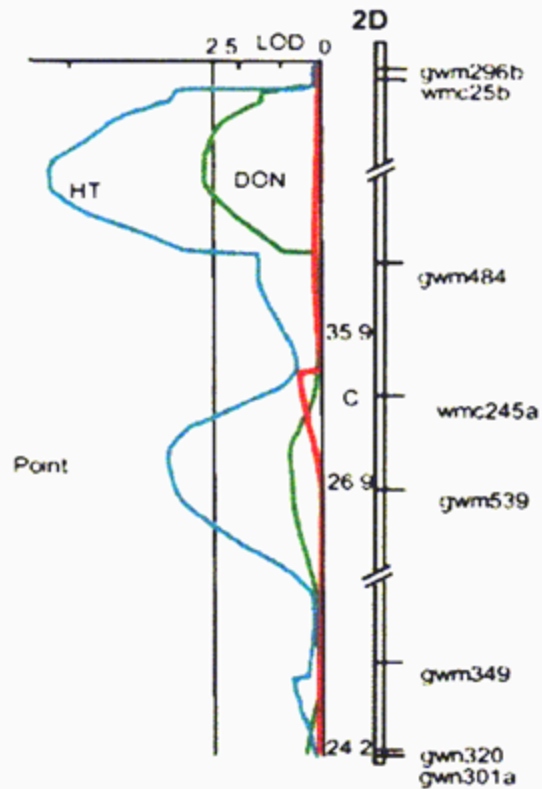
Line	FHB Index (%)	DON (ppm)
MSB 16	17.7	37.7
MSB 55	9.0	10.8
Odessa-129-2	7.8	9.6
Superb	46.3	47.2
Sumai 3	2.6	3.9

Markers assignments to FHB resistance QTLs obtained from alien sources

Source of resistance	Chromosome location	Markers	References
<i>T. monococcum</i> (A)	5A (M321)	wmc 705	--
	3A (Frontana)	gwm5	--
<i>Ae. cylindrica</i> (CD)	1D	cf65	--
	4D	wmc457, wmc622	--
	5D	gwm 292	--
	6D	barc 96	--
<i>T. carthlicum</i> (AB)	6B	wmc 397	Somers et al., 2006
	2B (Strongfield)	gwm 55	Somers et al., 2006
	1A	cfe2153	Singh et al., 2008
<i>T. timopheevii</i> (AG)	5A (Tc 67)	Cfa 2185	Malhipour et al., 2008

Pyramiding Sources of FHB Resistance





Wuhan/NuBay
Mapping Population
91 DH lines

FHB index and DON contents of 15 WS lines with their parents Sumai3 and Snowbird and 6 checks in the field FHB nursery of 2009, 2010 and 2011

Line	2009		2010		2011		Average		QTLs
	Index (%)	DON (ppm)	Index (%)	DON (ppm)	Index (%)	DON (ppm)	Index (%)	DON (ppm)	
Snowbird	36.67	14.30	36.67	31.23	41.67	7.20	38.34	17.58	x, x, x
Sumai3	3.00	0.67	30..	1.50	2.17	0.77	2.72	0.98	3B,5A,6B +
WS-44	3.33	1.33	3.00	2.73	3.50	0.53	3.28	1.53	3B, x, 6B +
WS-84	11.33	8.77	5.67	11.17	4.67	4.47	7.22	8.13	3B, x, 6B +
WS-131*	3.33	2.67	3.00	9.30	4.17	3.63	3.50	5.20	x, 5A, 6B +
WS-175	8.00	2.20	2.67	3.03	2.43	1.20	4.37	2.14	x, 5A, 6B +
WS-246	30.00	8.67	6.00	8.70	27.17	5.13	21.06	7.50	3B,5A, x -
WS-321*	15.65	5.67	3.00	8.67	10.93	5.23	9.87	6.52	3B, x, 6B +
WS-385	15.67	4.77	8.67	6.50	3.83	2.70	9.39	4.66	x, 5A, 6B +
WS-420	31.33	8.80	5.67	28.03	11.67	3.60	16.22	13.48	3B,5A, x H
WS-447	16.00	6.50	3.33	15.67	2.50	2.07	7.28	8.08	3B,5A, x -
WS-481*	3.00	0.53	2.67	28.67?	1.83	1.30	2.50	10.17	3B, x, 6B H
WS-630	21.00	6.80	7.00	16.80	3.50	4.67	10.50	9.42	3B,5A, x H
WS-745	14.67	5.63	3.67	4.97	3.67	3.43	7.34	4.68	3B,5A, x H
WS-795	3.00	4.27	7.67	8.70	30.17	7.43	13.61	6.80	3B, x, 6B +
WS-830	8.67	8.33	7.67	7.17	35.00	7.83	17.11	7.78	x, 5A,6B H
WS-886	7.00	2.07	8.33	3.13	2.10	1.07	5.81	2.42	3B,5A,6B +
ACS53619	33.33	17.63	61.67	15.47	66.67	9.47	53.89	14.19	x, x, x
AC Vista	90.00	35.93	91.67	28.10	98.33	19.57	93.33	27.87	x, x, x
Snowstar	-	-	43.33	18.47	56.67	3.93	50.00	11.20	x, x, x
AC Formost	-	-	98.33	22.80	100.00	13.60	99.17	18.20	x, x, x
AC Barrie	-	-	21.67	3.20	26.67	4.40	24.17	3.80	?
FL62R1	-	-	4.00	1.87	2.67	1.53	3.33	1.70	?



Spike symptoms of: 1) Roblin, 2) P1131-5, 3) P1142-1-2-6, 4) P1142-2-9, 5) P1142-2-5, 6) P1142-2-15, 7) P1142-3-B, 8) P1142-3-E, 9) P1142-3-1-5 at 21 days after inoculation with *Fusarium graminearum* spores

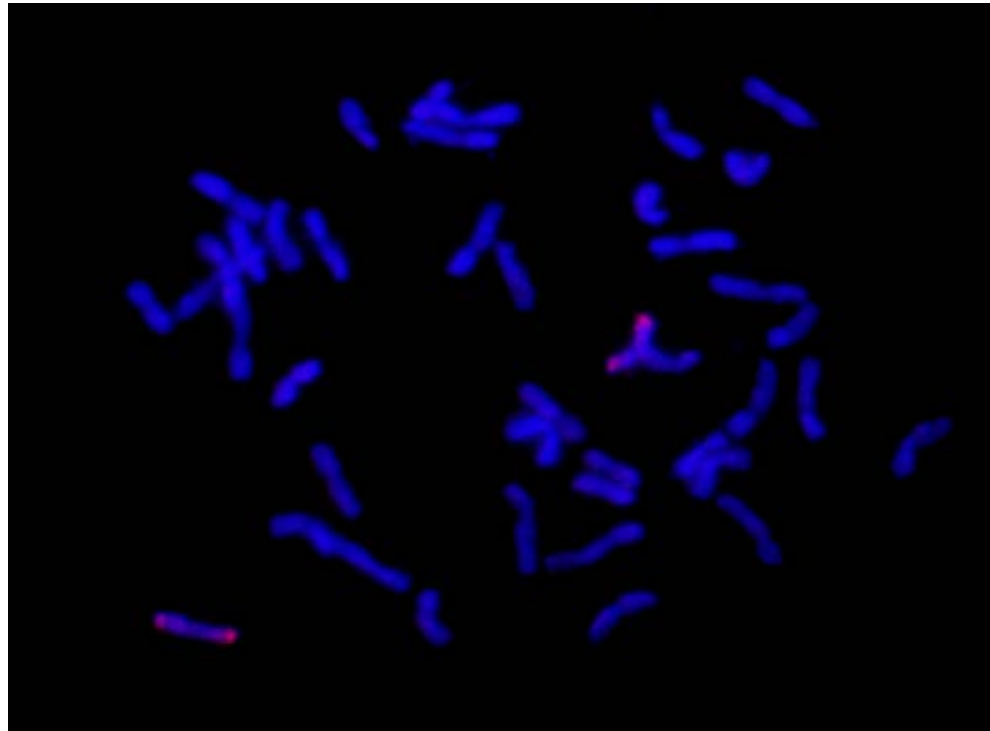
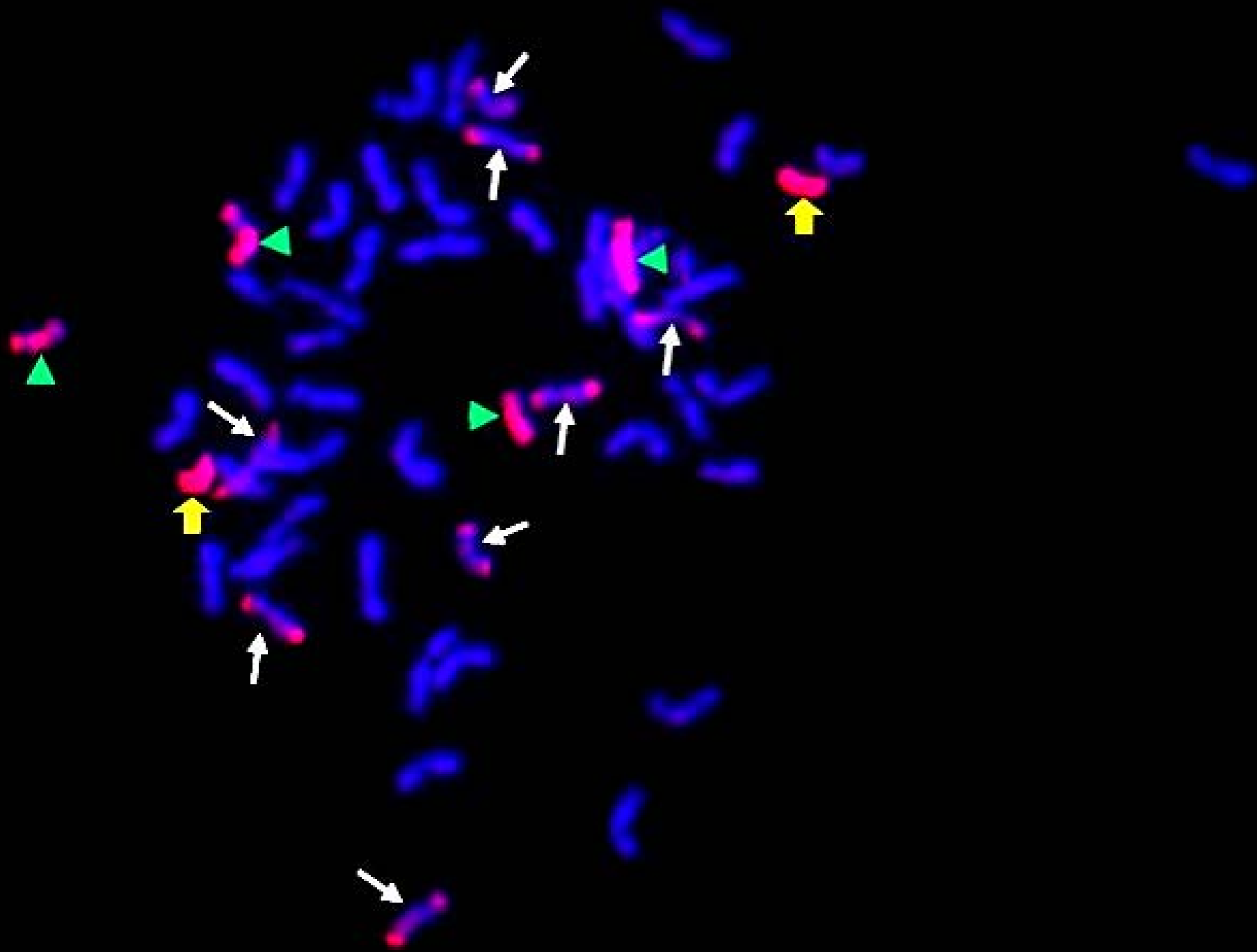


Fig. 3. P1142-3-1 ($2n = 42$) with two terminal translocation chromosomes (white arrows).



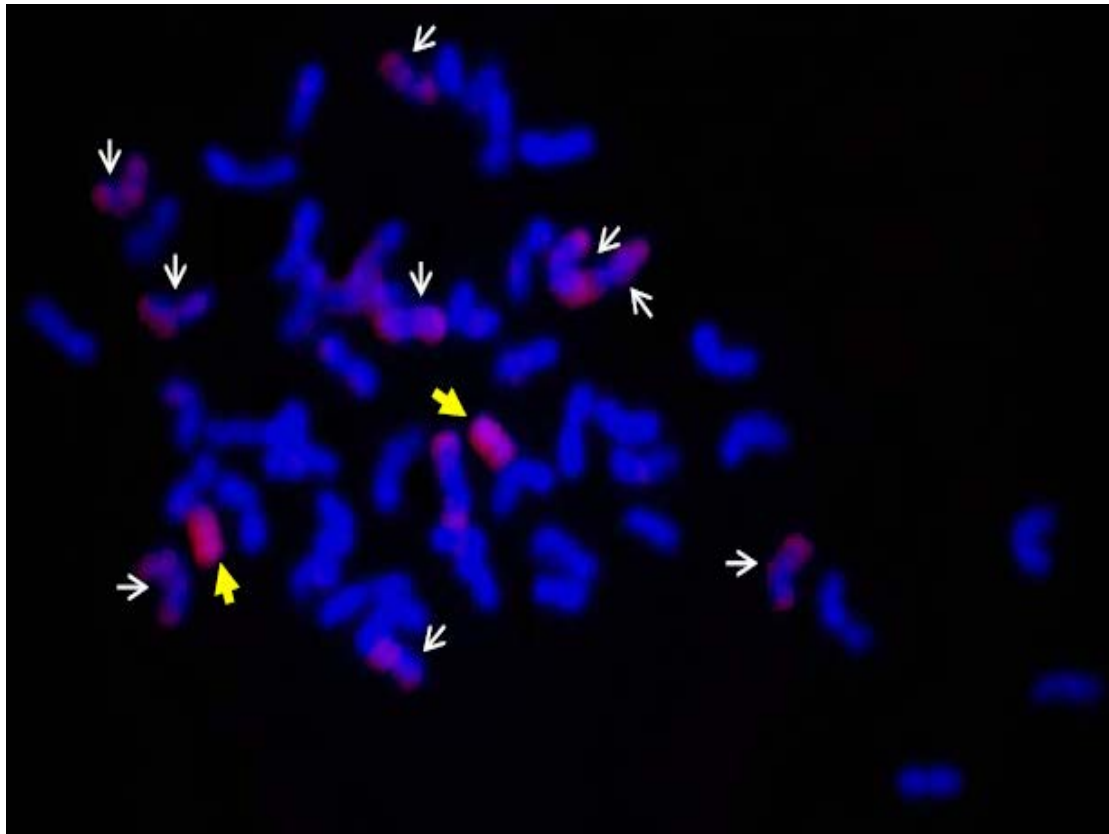


Fig. 2. P1142-2 ($2n = 54$) with 10 terminal translocation chromosomes (white arrows) and two entire chromosomes from *E. repens* (yellow arrows).

FHB symptoms on spikes of *Tritordeum* (durum x *Hordeum bogdanii* - ABH genomes) lines at 21 days after inoculation.



Roblin

Tritordeum

FHB symptoms and DON content in progenies of interspecific crosses between AC Strongfield and strains of *Tritordeum* (ABH) (field data 2005)

Generation	Number of lines	FHB Index (range)	DON Content (range) ppm
BC ₃ F ₄	27	0.5-10.5	0.8-5.5 7.4-11.9
Check			
AC Strongfield	–	6.8	16.5

FHB symptoms on
Tritordeum X AC
Strongfield
derivative



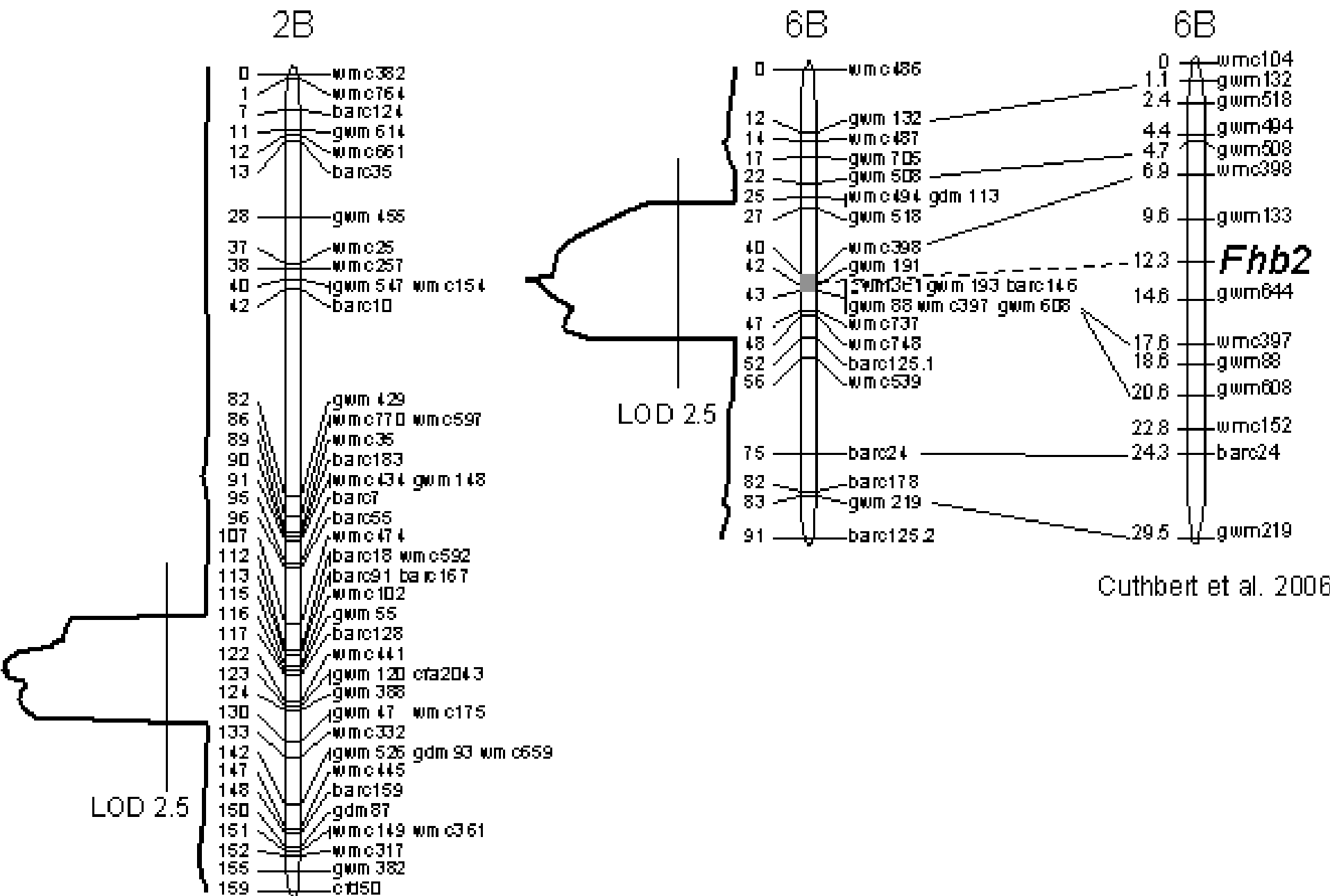
Trito-1-34

AC Strongfield

- Resistant accession identified.
- Mapping population of 100 RIL developed from cross to AC Strongfield



***Triticum carthlicum* (AB genome)**



FHB symptoms on a spike of
Triticale TMP16315 at 21
days after point inoculation.



TRITICALE FHB MAPPING

- TMP16315 x AC Ultima
- 100 DH lines
- Phenotyped at three locations

Infected spikelet of
Triticale at 21 days after
inoculation



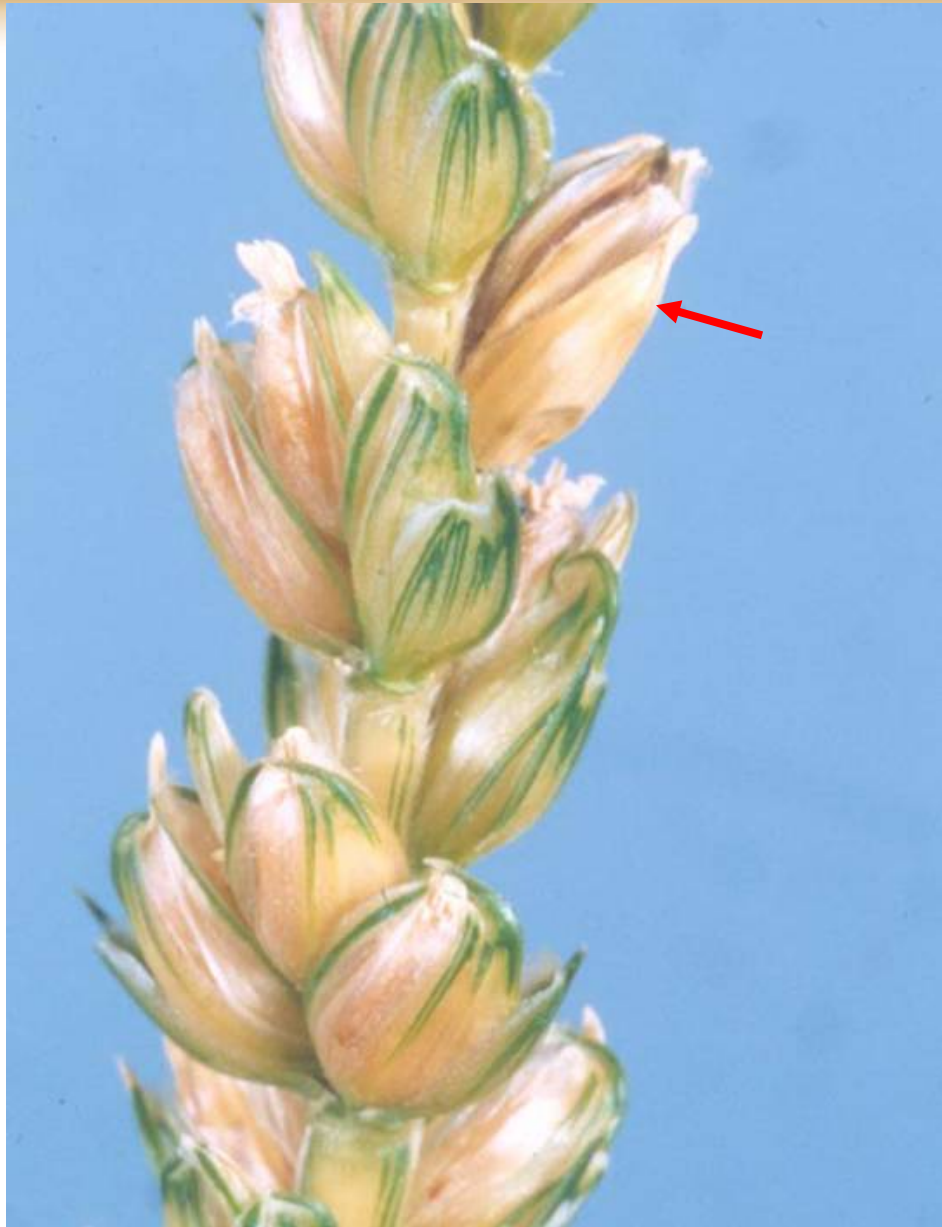
FHB Symptoms and DON Levels in Triticale lines(2 years average)

Line	Incidence(%)	Severity(%)	DON(ppm)
TMP16315	17.5	12.5	4.1
AC Ultima	6.0	47.5	17.5
Sumai3	5	5	1.2

FHB symptoms at
21 days after
point inoculation



Thinopyrum elongatum (E
genome, $2n=14$)



CS – 7E addition



Roblin



8801



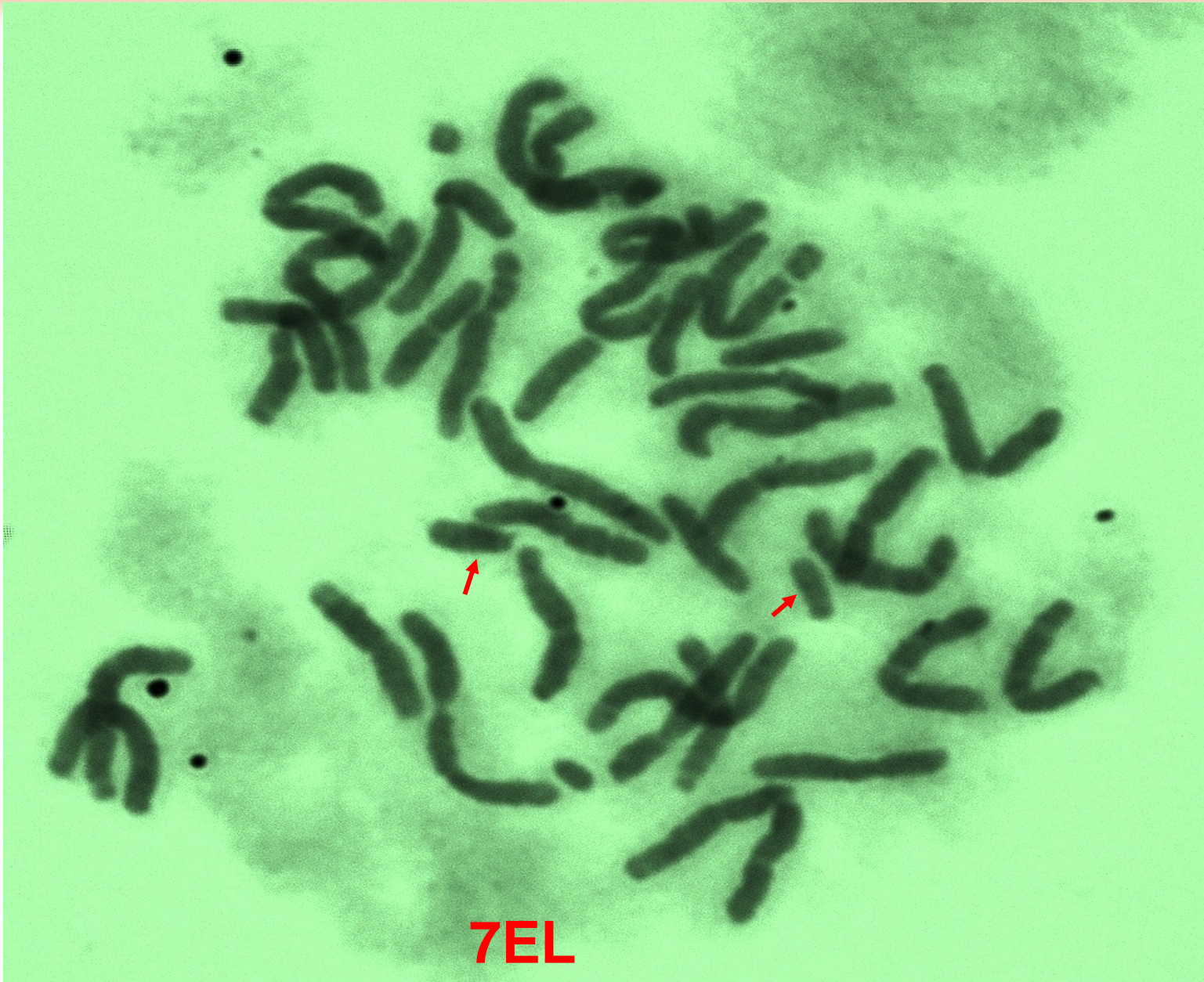
8802

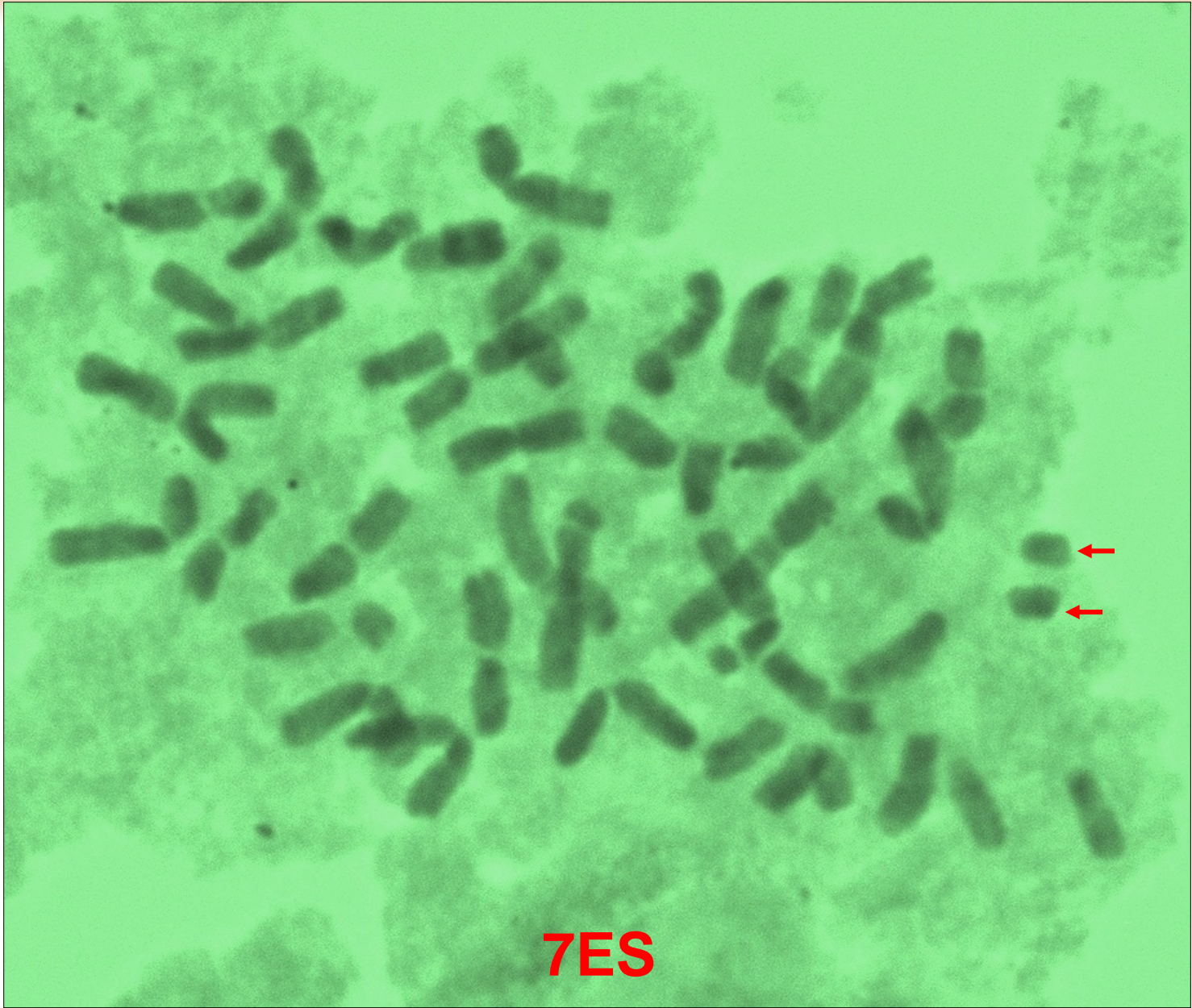


8803

FHB symptoms on 7E lines following point inoculation

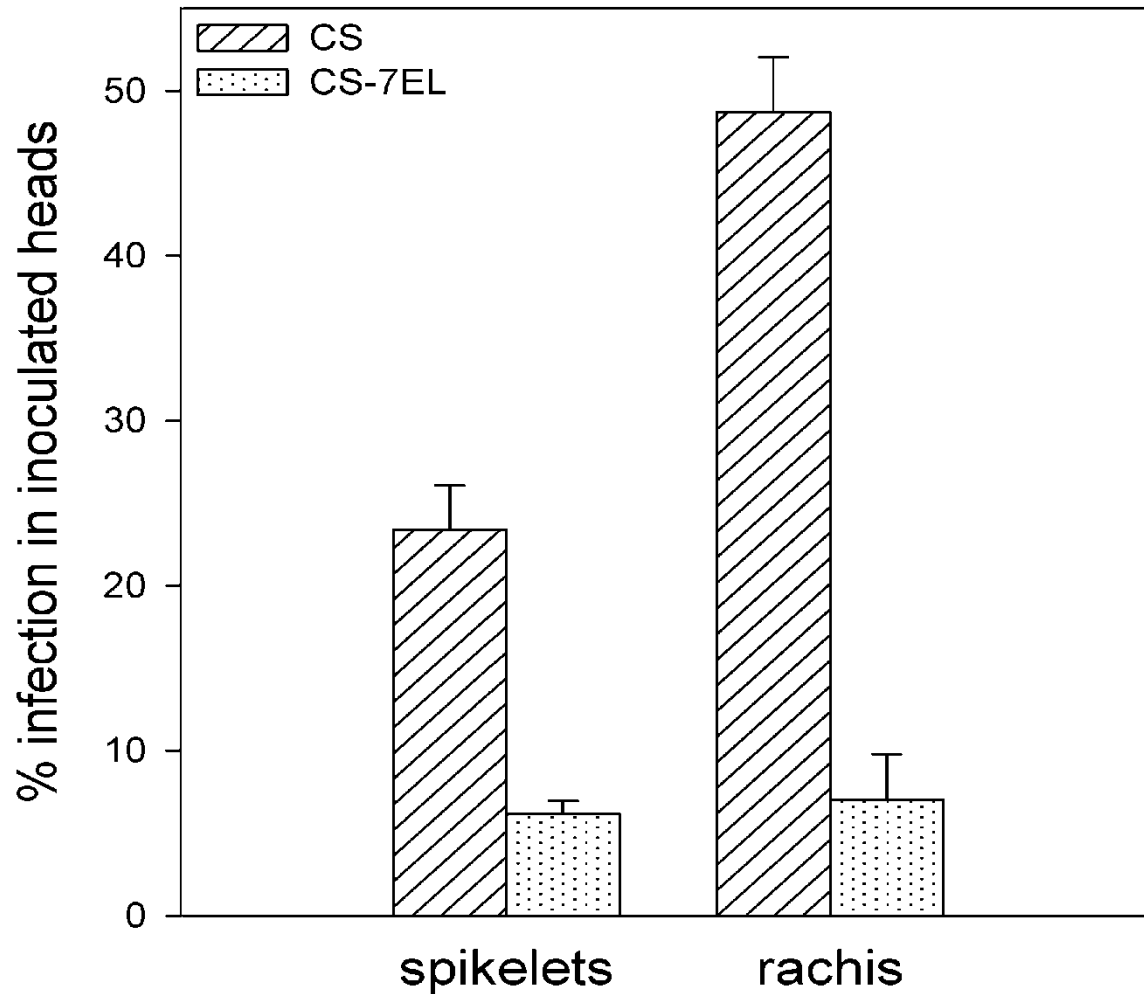
Aneuploid Lines	Severity % infected florets
7EL disomic addition $2n = 42 + 2t$	9.6
7ES disomic addition $2n = 42 + 2t$	86.5
7E disomic addition $2n = 44$	14.5
7E(7A) disomic substitution $2n = 42$	18.8
7E(7B) disomic substitution $2n = 42$	5.8
7E(7D) disomic substitution $2n = 42$	8.0
Chinese Spring parent	57.5
Sumai 3	2.8



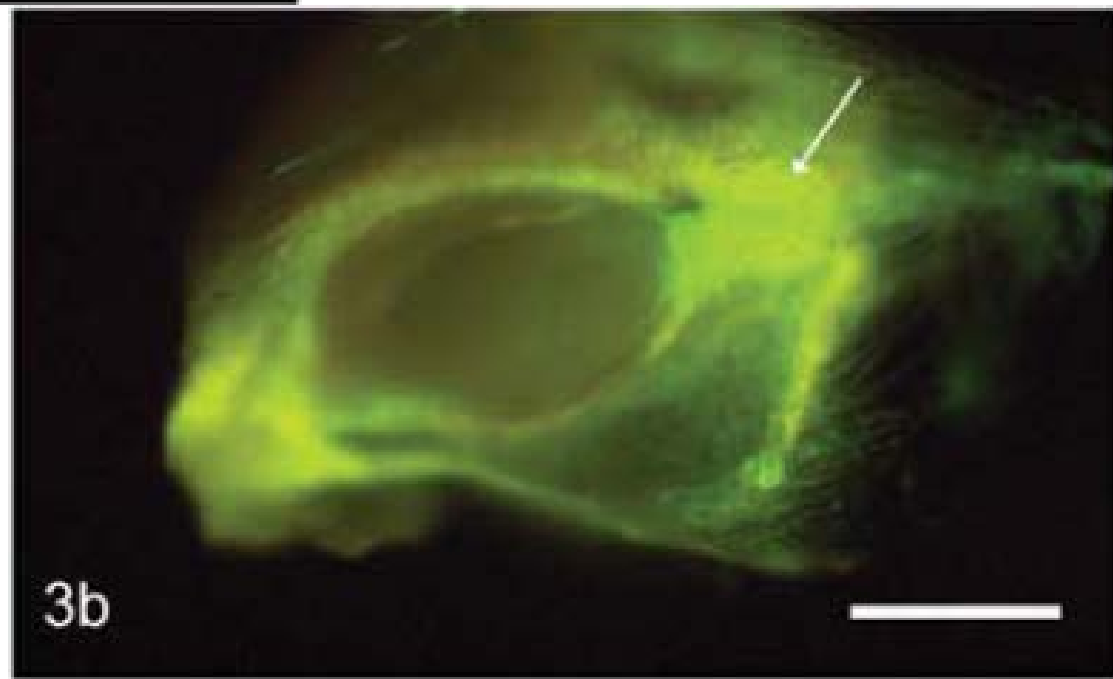
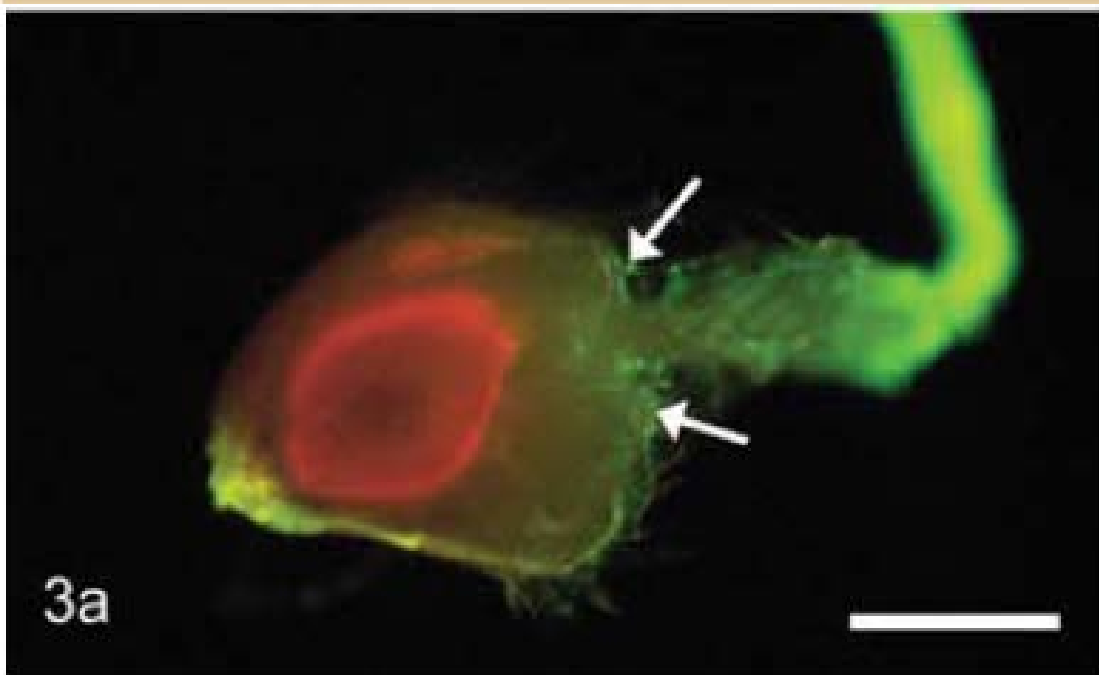


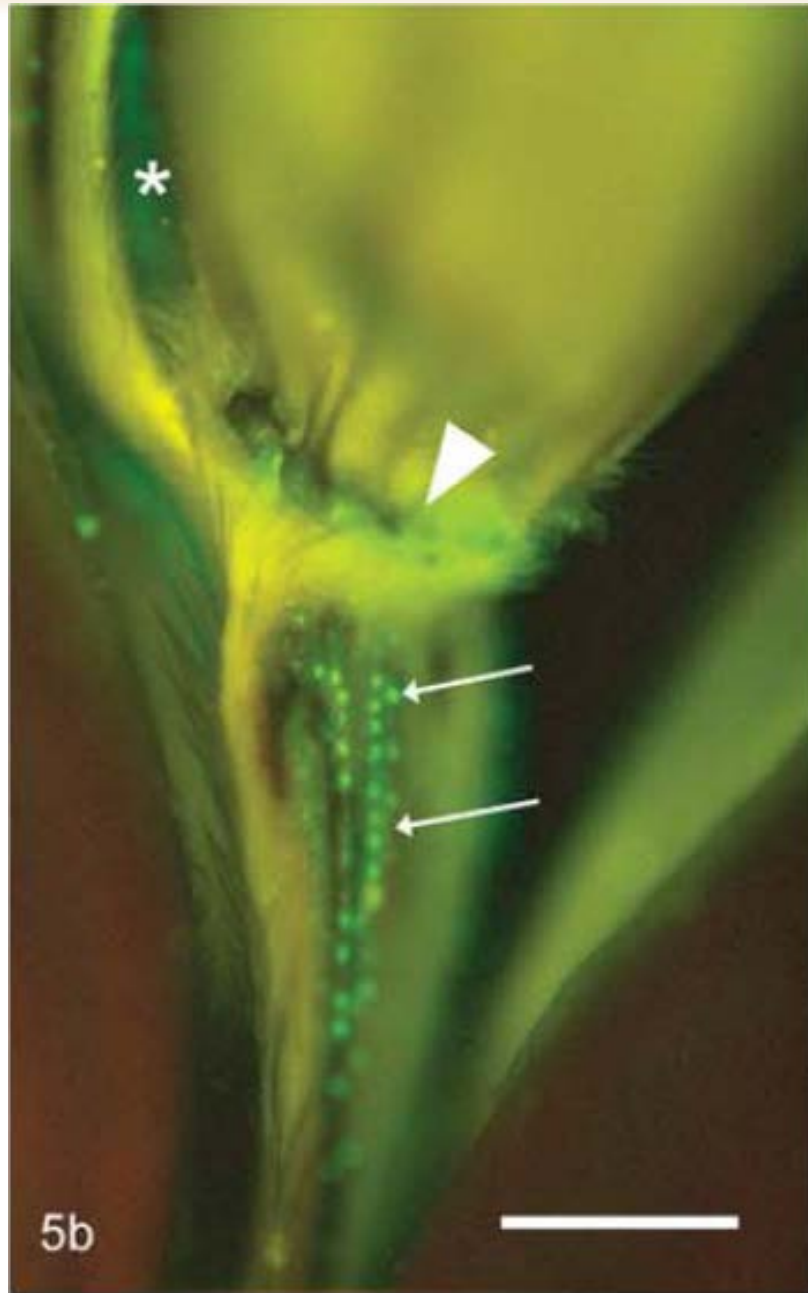


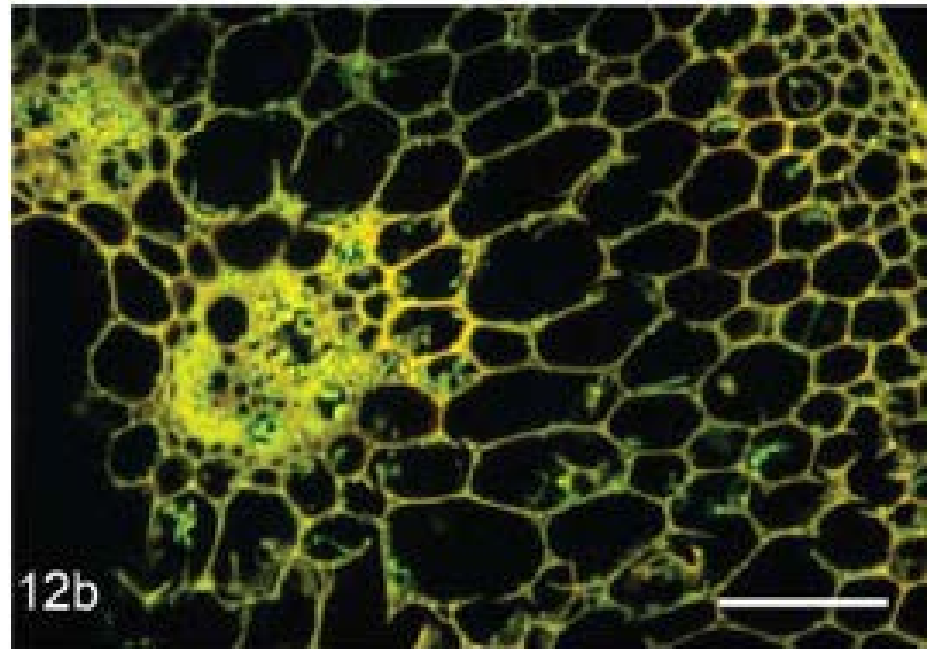
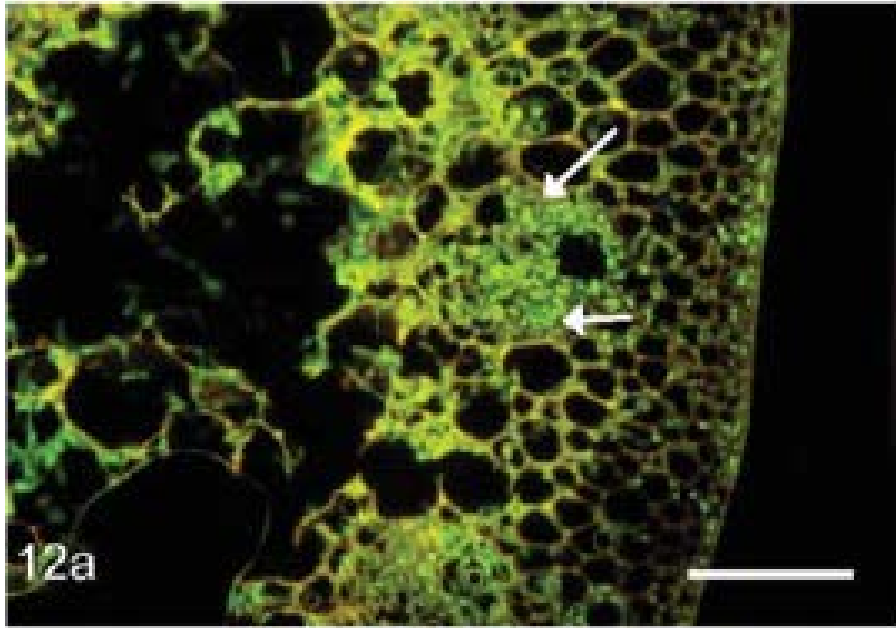
Spikes of CS (a) and CS-7EL(b) 4 days after inoculation. Arrows indicate inoculated spikelets. c,d) Rachis with florets removed above and below point of inoculation, 21 days after inoculation. c) CS, d) CS-7EL.

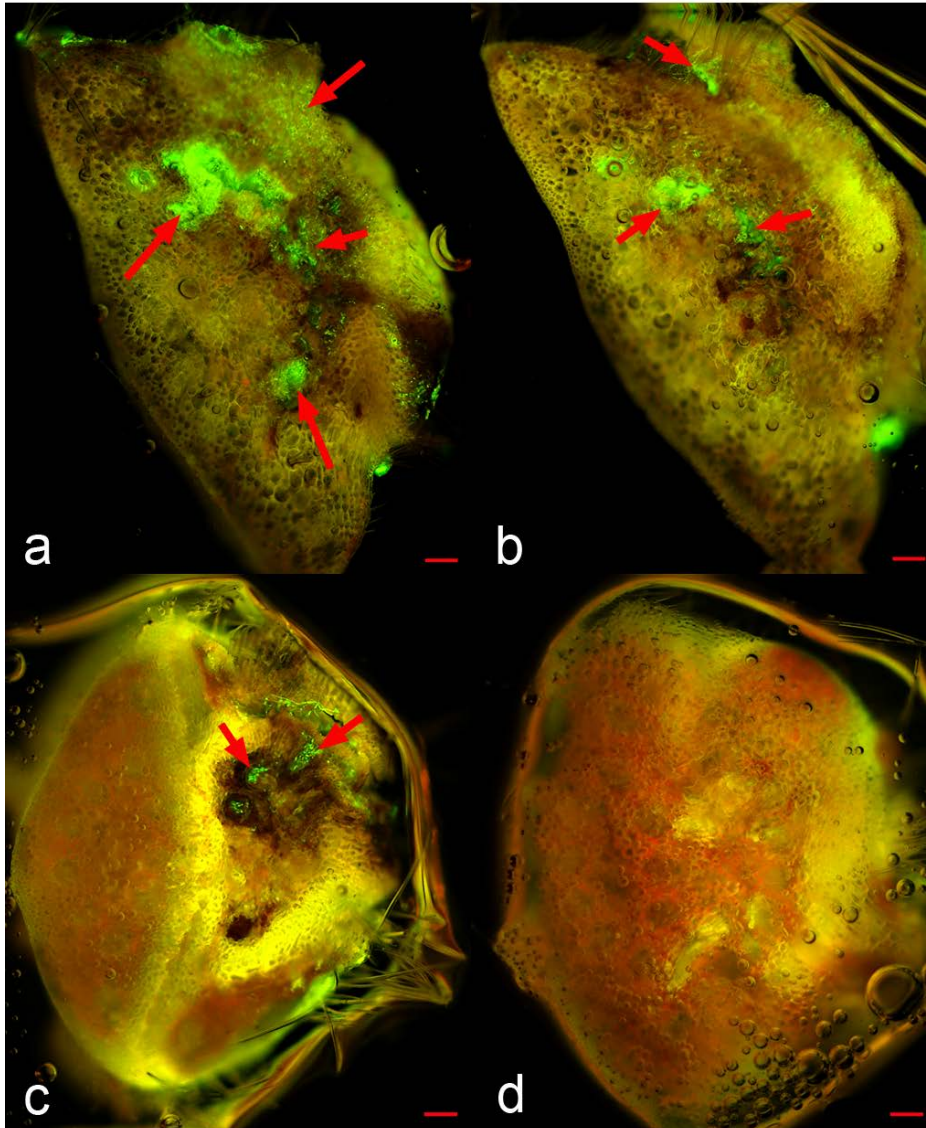


Average disease ratings for CS (n=19) and CS-7EL (n=23) inoculated heads, at 21 dpi. Inoculations were done with a suspension of 50,000 spores/ml.

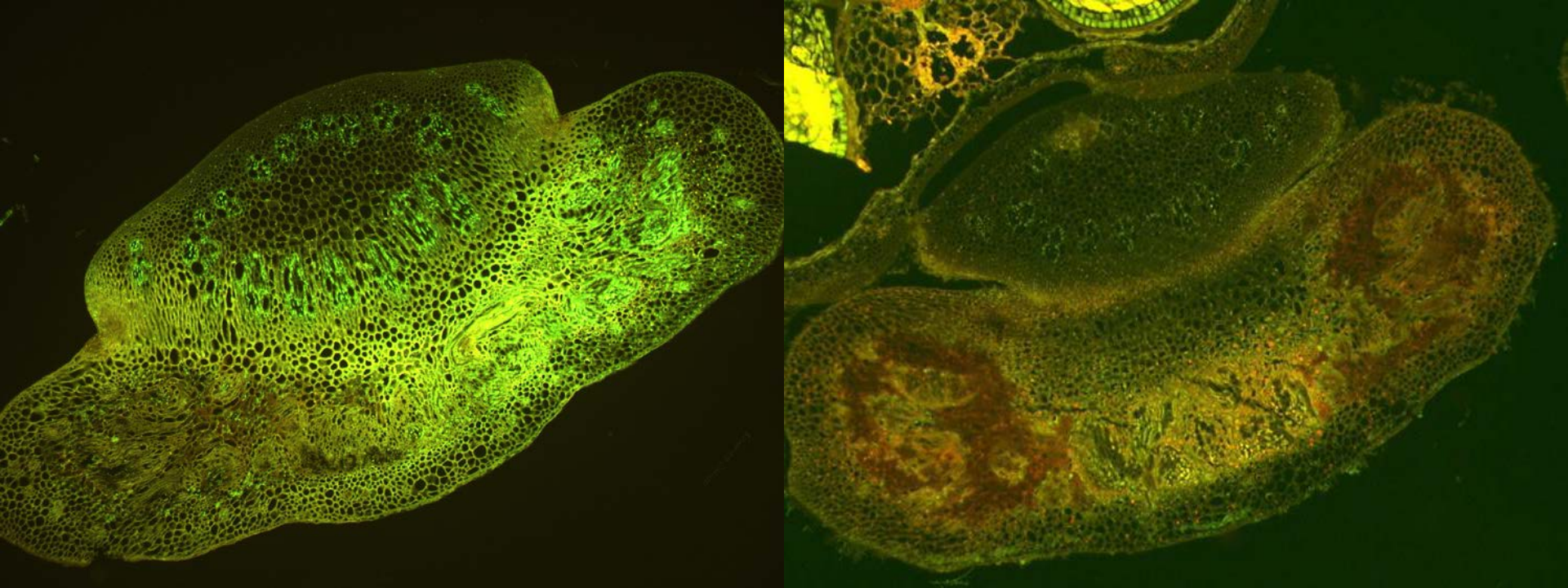








Hand sections through the node bearing the inoculated floret at 6 dpi. a) CS, top of the node (closest to point of inoculation) b) CS, bottom of the same node section, c) CS-7EL, top of the node (closest to point of inoculation) d) CS-7EL, bottom of the same node section. Bars = 200 μ m.

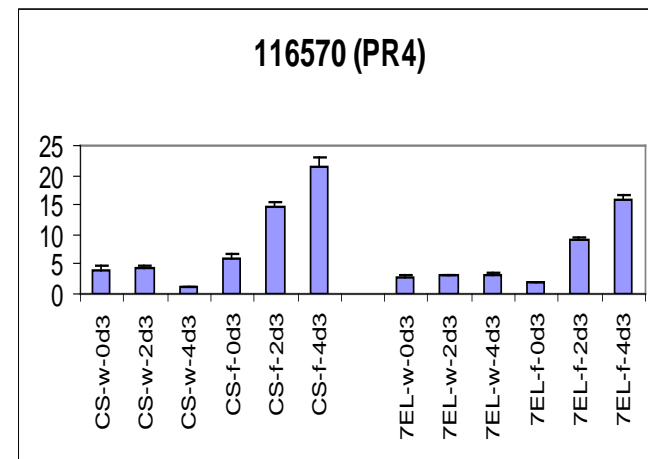
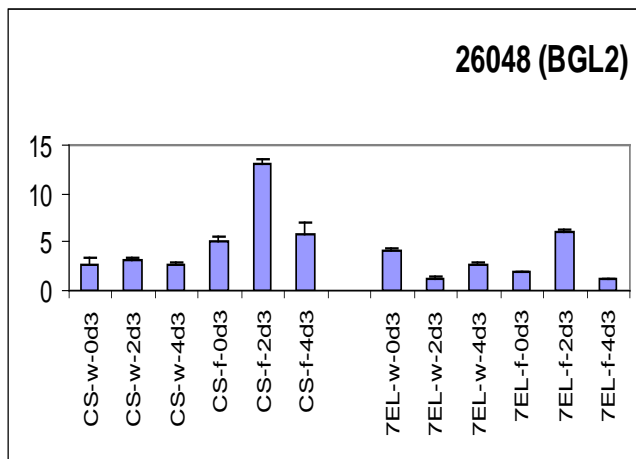
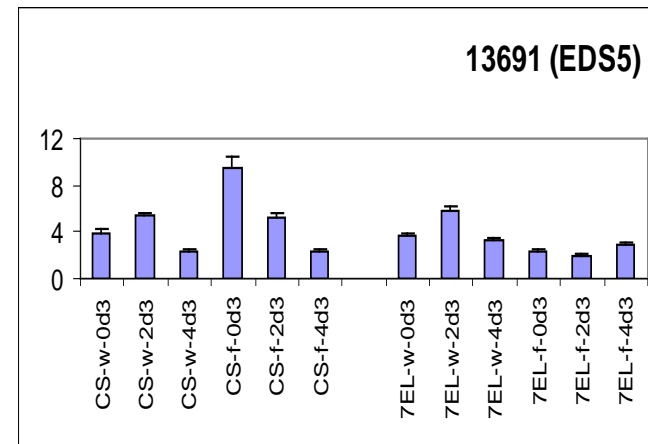
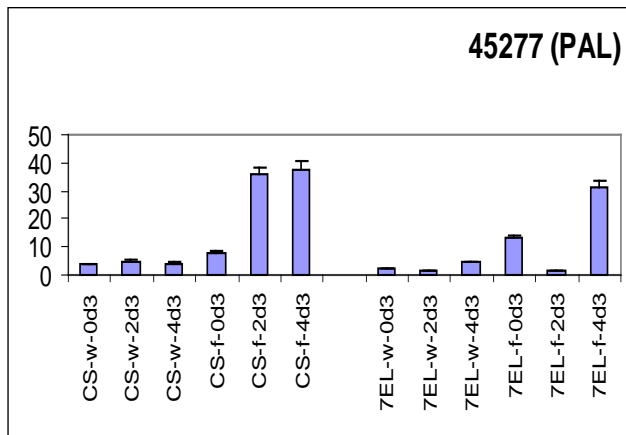


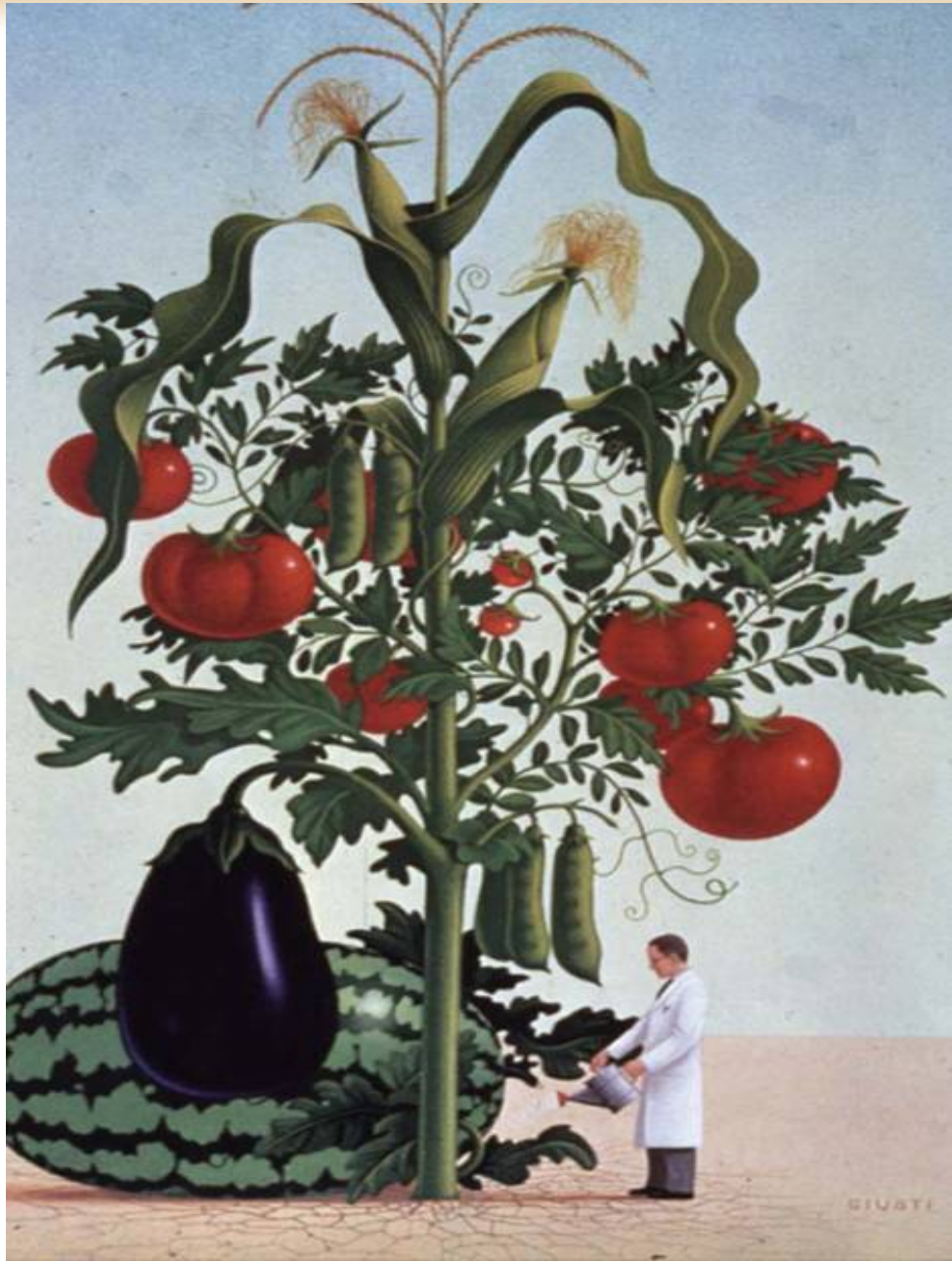
Chapais

Chevron

Barley. Node at base of inoculated floret, 11 days post inoculation

Comparison of gene expression profiles of selected defence response genes between CS and CS-7EL by RT-qPCR analysis.











Agriculture and
Agri-Food Canada

Agriculture et
Agroalimentaire Canada



Thank You

Canada 