

# **BREEDING FOR FUSARIUM MYCOTOXIN RESISTANT WHEAT GERMPLASM AT CIMMYT, MEXICO**

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# Growing concerns on Fusarium head blight globally

FHB is a major disease worldwide, causing not only yield loss, but also food and feed contamination

**Climate change** is likely to be increasing the risk of FHB epidemics

**Zero and minimum tillage practices** are associated with higher FHB severity where wheat is grown after maize



# FHB resistance components

- Type I for initial infection and Type II for fungal spread are the most important FHB resistance components
- Field FHB severity (or index) after spray inoculation represents a combination of Type I and Type II resistance
- DON content (for Type III) and Fusarium damaged kernels (FDK, for Type IV) are usually correlated with field FHB index, but generally moderate correlation is observed
- Field FHB index is often associated with days to heading (DH), plant height (PH) and anther extrusion (AE)



# FHB resistance genes

FHB genes	Source of resistance	Chromosome	Resistance Type	Flanking markers	References
<b><i>Fhb1</i></b>	Sumai 3	3BS	II	Perfect markers	Rawat et al. 2016
<b><i>Fhb2</i></b>	Sumai 3	6BS	II	gwm133 and gwm644	Cuthbert et al. 2007
<b><i>Fhb3</i></b>	<i>Leymus racemosus</i>	T7AL-7Lr#1S	II	BE586744-STS, BE404728-STS, and BE586111-STS	Qi et al. 2008
<b><i>Fhb4</i></b>	Wangshuibai	4BL	I	barc20 and wmc349	Xue et al. 2010
<b><i>Fhb5</i></b>	Wangshuibai	5AS	I	barc56 and barc100	Xue et al. 2011
<b><i>Fhb6</i></b>	<i>Elymus tsukushiensis</i>	1Ets#1S	II	wg1S_snp1	Cainong et al. (2015)
<b><i>Fhb7</i></b>	<i>Thinopyrum ponticum</i>	7DS.7el2L	II	sdauK66 and cfa2240	Guo et al. (2015)



# CIMMYT experimental stations in Mexico



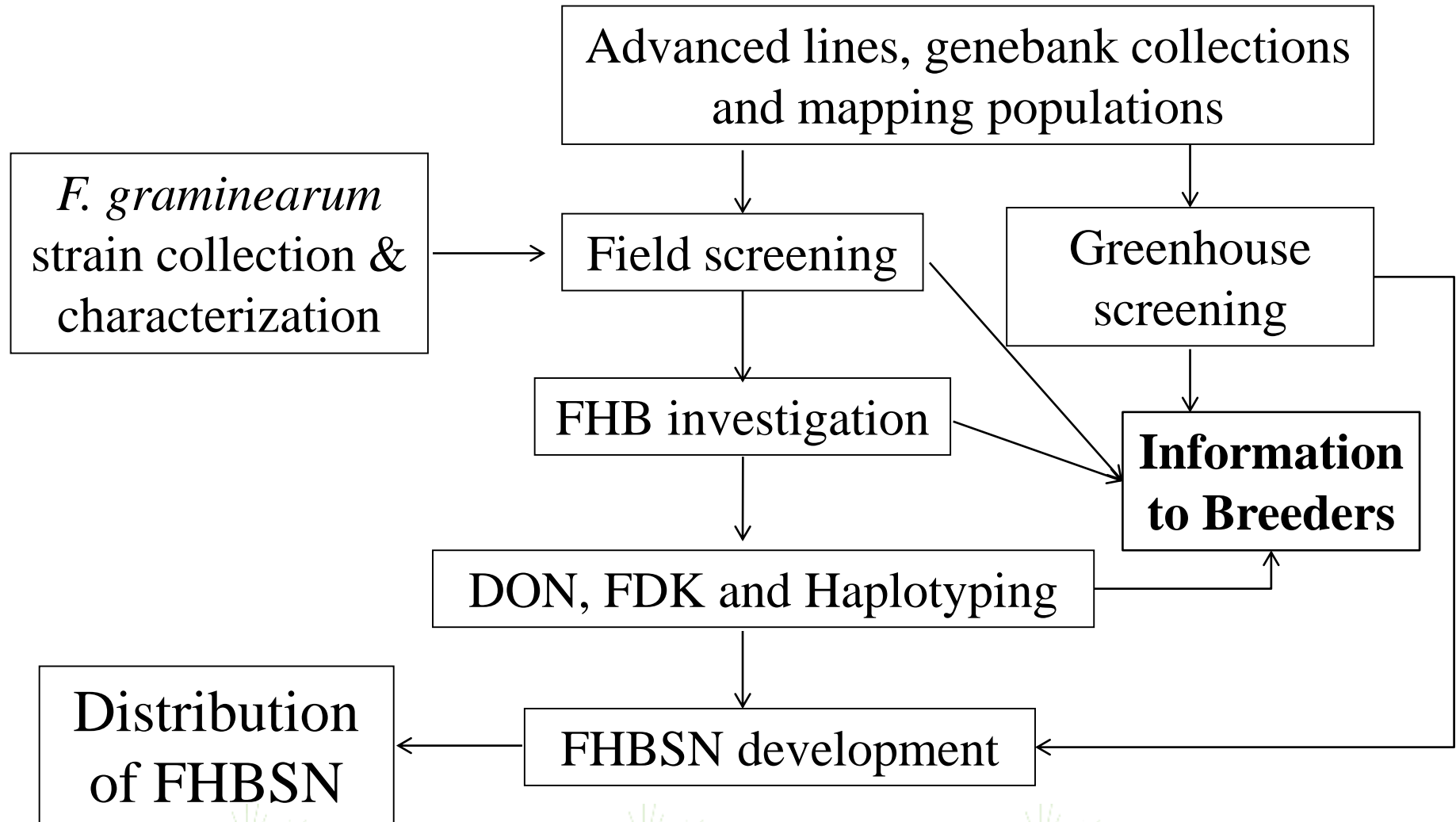


# Field Screening: CIMMYT, El Batan (since 2006)

~ 1.9 hectares



# Workflow of the FHB research at CIMMYT





## List of some entries from the 15<sup>th</sup> FHBSN

Entries	Mexico (El Batán )								International locations				
	FHB 2010	FHB 2011	DON 2011	FHB 2012	DON 2012	FHB 2013	DON 2013	FDK 2013	NO 2013	UY 2013	NE 2013	CA 2013	JP 2014
6340362	12.7	4.9	0.6	1.3	0.2	13.4	3.0	2.5	16.6	6.0	55.0	10.0	18.0
6000734	14.3	2.9	1.3	3.2	0.5	24.6	6.7	2.5	43.2	15.0	37.0	10.0	15.0
6000696	2.4	0.5	0.3	3.5	0.9	20.8	2.2	3.0	37.9	2.0	10.0	10.0	7.0
6342108	12.5	3.9	1.8	3.0	0.9	20.8	4.4	2.0	22.9	2.0	37.0	30.0	13.0
6340565	8.9	2.7	0.4	2.7	1.0	20.3	1.7	3.5	19.3	14.0	17.0	10.0	6.0
6340604	8.9	4.1	2.9	1.7	2.4	23.9	5.3	4.0	28.4	0.3	65.0	50.0	24.0
6340966	11.3	4.4	0.7	3.3	0.5	4.3	2.4	4.0	27.7	1.0	17.0	10.0	11.0
6000673	14.3	1.4	1.3	1.8	1.0	11.0	6.0	2.0	50.0	8.0	37.0	30.0	25.0
6176474	8.6	1.8	1.2	3.1	1.4	15.2	6.0	1.5	24.8	10.0	25.0	10.0	52.0
6342246	4.3	3.1	0.6	3.1	0.7	12.7	3.6	2.5	23.9	7.0	55.0	50.0	6.0
6340672	2.1	3.4	1.0	1.8	1.5	13.1	5.3	2.0	22.1	18.0	37.0	50.0	9.0
6340845	7.3	4.3	1.3	1.8	0.4	10.9	2.7	1.5	35.9	21.0	60.0	10.0	12.0
6342263	8.1	3.9	0.9	2.7	1.1	13.9	6.0	2.0	38.9	0.3	55.0	10.0	51.0
6342266	8.2	3.8	0.2	2.5	0.5	8.5	2.3	2.0	35.2	5.0	50.0	50.0	9.0
6340765	7.4	2.9	1.5	2.6	0.7	21.8	3.8	3.0	29.8	8.0	17.0	30.0	13.0
6343651	17.9	2.7	0.9	1.4	0.7	16.0	2.6	2.5	19.1	18.0	50.0	30.0	9.0
6342336	6.5	1.6	1.4	2.8	2.2	7.7	5.0	2.5	45.0	1.0	25.0	50.0	21.0
6342353	18.7	3.3	0.6	1.1	0.5	11.8	1.3	4.5	48.2	18.0	65.0	10.0	13.0
6343684	18.6	4.5	1.0	3.0	0.7	5.8	1.6	2.5	35.9	16.0	60.0	10.0	12.0
6340862	11.1	4.2	0.7	1.7	0.5	11.3	2.6	1.5	32.9	1.0	50.0	10.0	3.0
6343369	16.4	3.8	1.3	2.7	3.1	15.9	5.9	1.5	22.6	21.0	60.0	10.0	25.0
6343743	7.3	4.3	0.6	0.6	0.7	12.9	2.4	2.5	34.0	12.0	17.0	10.0	13.0
5999852	1.1	2.4	2.5	2.7	0.9	18.9	3.2	4.5	59.1	9.0	37.0	10.0	25.0
Sumai 3	1.5	1.4	0.1	1.0	0.0	1.0	0.3	1.0	7.6	1.0	3.0	10.0	7.0
Gamenya	68.0	78.7	3.9	72.0	7.5	60.4	6.1	8.5	44.6	24.0	70.0	70.0	65.0

Osman et al. (2015) Phenotypic and genotypic characterization of CIMMYT's 15<sup>th</sup> international Fusarium head blight screening nursery of wheat. *Euphytica* 205:521-537.



# FHB markers used in CIMMYT's haplotyping system

Source of resistance	Chromosome	Resistance Type	Flanking marker(s)	Locus
Sumai 3	3BS	II	Functional markers	<i>Fhb1</i>
	5AS	I	<i>barc186</i> and <i>barc180</i>	<i>Fhb5</i>
	6BS	II	<i>gwm133</i> and <i>wmc179</i>	<i>Fhb2</i>
Frontana	3A	I	<i>dupw227</i>	
	5AS	I	<i>barc197</i> and <i>gwm129</i>	<i>Fhb5</i>
Wuhan 1	2DL	II	<i>wmc144</i> and <i>wmc245</i>	
	4BS	II	<i>wmc238</i> and <i>gwn149</i>	<i>Fhb4</i>
CJ 9306	2DL	II	<i>gwm157</i> and <i>gwm539</i>	
<i>T. dicoccoides</i>	3A	II	<i>gwm2</i>	
	7A	II	<i>barc121</i> and <i>wmc488</i>	



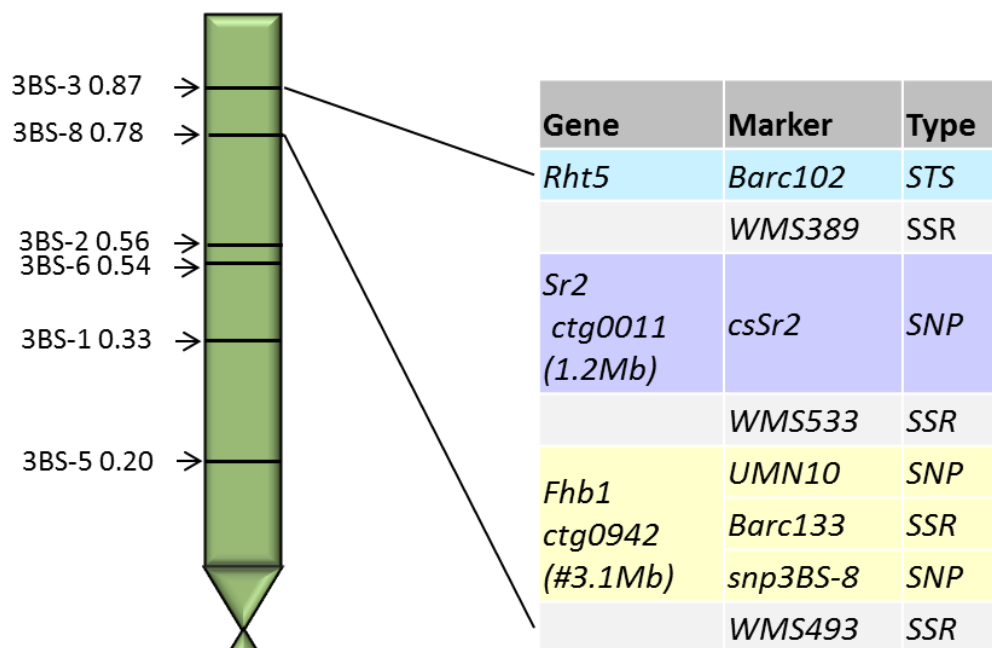
## Prediction on frequency (%) of FHB resistant genes/QTLs for FHBSN entries

<b>FHBSN</b>	<b><i>Fhb1</i></b> (Type II-3BS)	<b><i>Fhb2</i></b> (Type II-6BS)	<b><i>Fhb4</i></b> (Type I-4BL)	<b><i>Fhb5</i></b> (Type I-5AS)	<b>2DLc</b> (Type I & II)	<b><i>Fhb7AC</i></b> (Type II)
13 <sup>th</sup>	0	6.3	0	6.3	56.3	12.6
14 <sup>th</sup>	0	0	6.7	6.7	66.7	6.7
15 <sup>th</sup>	0	4.5	54.5	27.3	11.4	36.4
16 <sup>th</sup>	0	0	0	NA	31.3	NA
17 <sup>th</sup>	0	2.6	5.2	NA	15.4	NA
18 <sup>th</sup>	0	0	4.2	0	2.1	20.8
19 <sup>th</sup>	22.9	0	16.7	0	9.3	12.5



# Development of *Fhb1*/*Sr2* recombinant lines

- *Sr2* and *Fhb1* are linked **in repulsive** in the *Sr2*/*Fhb1* region of *wms533* and *wms493* on 3BS.
- Estimated at 5 cM (Bernardo et al. 2012) or 2.4 cM apart (Zhang et al. 2016)



# Development of *Fhb1/Sr2* recombinant lines

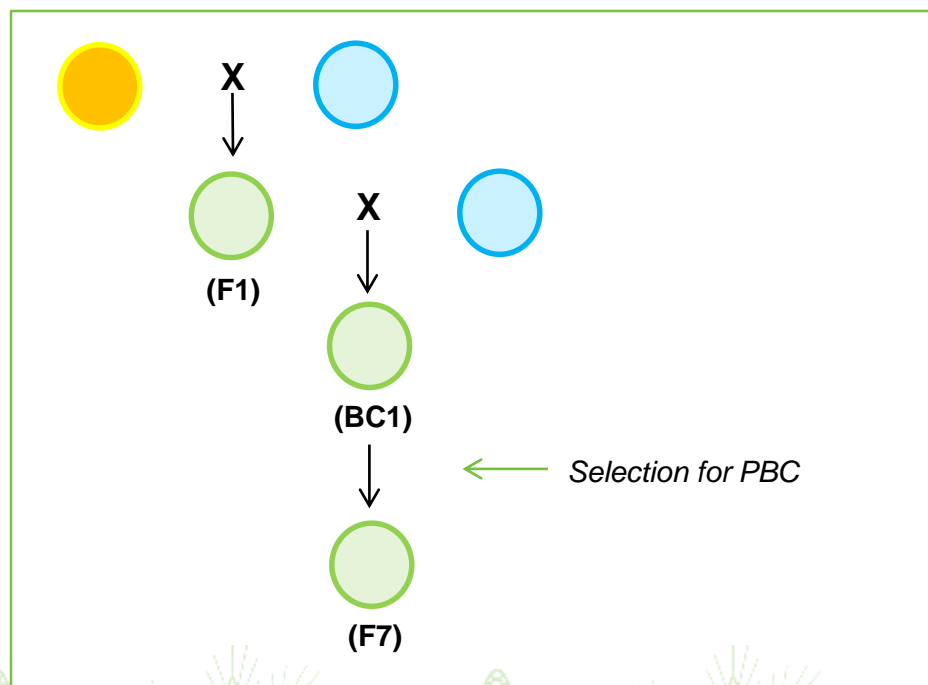
**Recombinants (*Sr2-Fhb1* in coupling) in the background of Hartog**

H-S A, H-S B, H-S C, H-S D



## CIMMYT advanced line

Vorobey, Roelfs 07, Munal #1, Quaiu #1, Navojoa 07, Super Seri 142,  
Guayacan Inia/Guanay//Porron\_4/Bejah\_7/3/Vanrrikse\_12/Snitan, Janz





# Development of *Fhb1*/*Sr2* recombinant lines

- Selection for PBC during population advance
- 2013: Genotyping of 19 F7-populations and parents (1,235 entries) with *Fhb1* and *Sr2* markers
- 2013: FHB evaluation of 234 lines in the field
- Summer 2014: FHB and FDK evaluation of 81 lines in the field  
Winter 2014: FHB Type II evaluation of 30 lines in the greenhouse
- 2015: FHB, FDK and DON evaluation of 30 lines in the field, additional evaluation for YR, TS, SB, STB, SNB
- 2016: Stem rust field nursery of 30 selected lines in Kenya; and the presence of *Fhb1* was confirmed with the newly developed diagnostic markers



# Characterization of the *Fhb1*/*Sr2* recombinant lines

Cross Name	FHB 2014	FHB 2015	FHB Typell	Spot blotch	STB	TS	YR	SR	PBC	Sr2	Fhb1		
H-S A/2*ROLF07	17.5	7.1	39.7	1037.0	996.0	560.2	MR	MSS	+	+	+	+	-
H-S A/2*VORB	12.0	0.5	8.4	669.8	438.9	601.2	R	MSS	+	+	+	+	+
H-S A/2*VORB	17.2	5.4	6.4	883.6	753.7	508.3	R	MSS	+	+	+	+	+
H-S A/2*MUNAL #1	16.3	7.3	9.4	680.6	801.2	480.9	MR	MSS	+	+	+	+	+
H-S A/2*QUAIU #1	26.9	9.7	11.1	1002.5	1338.6	472.8	R	S	+	+	+	+	+
H-S A/2*QUAIU #1	13.5	4.0	14.9	427.8	1515.1	617.9	R	M	+	+	+	+	+
H-S B/2*NAVJ07	19.2	19.3	2.8	330.6	1067.0	362.3	MS	S	+	+	+	+	+
H-S B/2*NAVJ07	10.8	9.3	13.9	756.2	1004.9	358.0	MR	M	+	+	+	+	+
H-S B/2*MUNAL #1	23.9	8.9	2.1	745.4	814.8	371.0	MR	M	+	+	+	+	+
H-S C/2*VORB	4.1	3.9	5.5	311.1	629.0	372.2	R	MSS	+	+	+	+	+
H-S C/2*VORB	19.2	2.2	14.4	220.4	652.2	580.6	R	MSS	+	+	+	+	+
H-S C/2*VORB	5.5	21.9	7.1	408.3	1036.7	525.0	MR	M	+	+	+	+	+
H-S C/2*MUNAL #1	21.9	4.5	7.8	792.9	1183.6	625.3	MR	M	+	+	+	+	+
H-S C/2*SUP152	42.0	17.2	4.4	NA	1033.0	714.8	R	M	+	+	+	+	+
H-S D/2*ROLF07	10.4	30.0	45.3	777.8	889.5	714.8	R	MSS	+	+	+	+	+
H-S D/2*ROLF07	4.4	25.4	11.8	410.5	668.5	701.9	R	MSS	+	+	+	+	+
H-S D/2*ROLF07	8.6	11.7	11.5	611.4	651.2	714.8	R	MSS	+	+	+	+	+
H-S D/2*VORB	13.8	5.7	0.0	805.9	493.5	474.4	R	M	+	+	+	+	-
H-S D/2*VORB	6.4	11.0	8.0	743.2	660.5	502.5	R	M	+	+	+	+	NA
H-S D/2*MUNAL #1	6.2	4.4	14.0	1140.7	873.5	673.8	R	MSS	+	+	+	+	+
H-S D/2*SUP152	31.3	4.4	9.6	1400.0	1133.3	560.2	R	M	+	+	+	+	+
H-S D/2*QUAIU #1	6.7	16.0	15.0	730.2	584.6	346.3	R	M	+	+	+	+	+
H-S B/2*NAVJ07	20.4	2.5	0.0	1037.0	1580.9	413.6	R	M	+	+	+	+	+
H-S C/2*VORB	8.1	2.5	12.6	665.4	588.9	456.8	R	MSS	+	+	+	+	+
H-S D/2*MUNAL #1	9.5	2.7	0.0	1160.2	829.0	608.6	MR	M	+	+	+	+	+
H-S B/JANZ-Rth5	14.2	3.0	11.7	1449.7	1728.1	158.0	S	S	+	+	+	+	-
H-S B/JANZ-Rth5	10.1	6.4	14.5	1566.4	1767.0	380.9	S	S	+	+	+	+	-
H-S B/JANZ-Rth5	7.2	15.0	9.3	1501.5	1688.3	158.6	S	M	+	+	+	+	-
H-S B/JANZ-Rth5	10.5	4.5	0.0	829.6	1679.6	378.1	S	M	+	+	+	+	-
R check	0.2	7.7	0.1	272.2	388.3	346.3	R						
S check	79.6	73.6	62.5	1002.5	1696.3	555.2	MS						

11 of the recombinant are included in the 19<sup>th</sup> FHBSN and was distributed globally in 2017

# Development of *Fhb1/Sr2* recombinant lines

- 11 of the recombinant are included in the 19th FHBSN and was distributed globally in 2017
- 7 of the recombinant lines have been incorporated in wheat breeding program since 2014, and F7 lines were evaluated in our FHB screening field, showing promising results
- Soon elite lines with *Fhb1/Sr2* will be available in future IBWSN and FHBSN nurseries
- The utilization of *Fhb1* will significantly contribute to the improvement of the Type II resistance of CIMMYT germplasm.



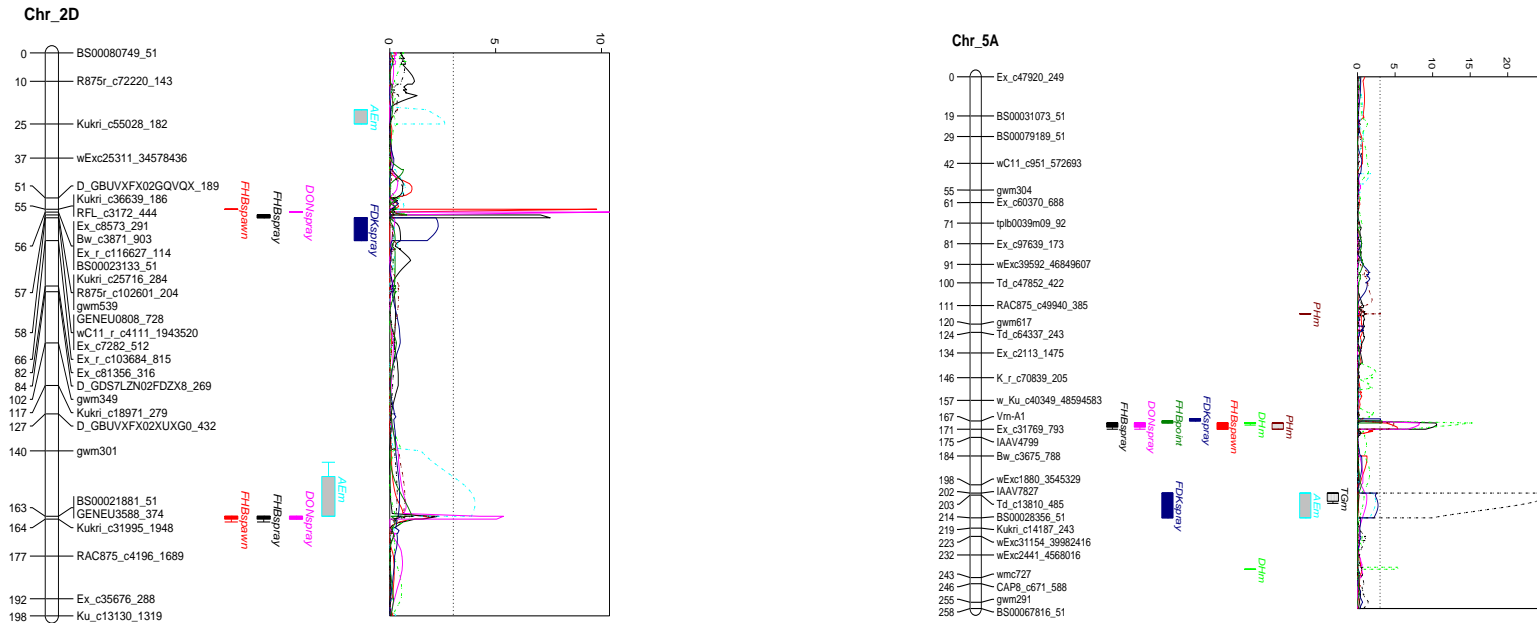
# QTL mapping for Soru#1

- CIMMYT FHB resistant line 'Soru#1' (SABUF/5/BCN/4/RABI//GS/CRA/3/AE.SQUARROSA (190), was hybridized with a German cultivar 'Naxos' to generate 131 recombinant inbred lines
- Phenotyping was carried out for FHB and associated traits in spray inoculated experiments in El Batan (Mexico, 2011-2012), spawn inoculated experiments in Ås (Norway, 2011-2012) and point inoculated experiments in Nanjing (China, 2011 and 2013), with two field trials at each location
- Genotyping was done with the Illumina iSelect 90K SNP wheat chip, along with selected SSR and STS markers





# QTL mapping for Soru#1



QTL	Position	Left marker	Right marker	Spray inoculation							Spawn inoculation			Source	Associated traits
				FHB index			FDK	DON content			FHB severity				
				2011	2012	Mean		2012	2011	2012	Mean	2011	2012		
2DLc	55.3-58.2	Kukri_c36639_186	Ex_c7282_512	18.1	15.8	15.9	7.4	20.1	13.6	16.5	15	22.4	18.2	S	
2DL	162.6-163.4	BS00021881_51	GENEU3588_374	6.3	8.6	4.4		9.4	7.5	6.1		5.3	3.2	N	AE
4AL	101.2-102.6	Ex_c11968_204	RAC875_c35979_263	7.1		5.4					6.8	4	5.9	S	DH
4DS	0.0-15.9	Rht-D1	D_c56766_278				19							N	PH
5AL	167.2-171.2	Vrm-A1	Ex_c31769_793	36.8	11.2	22.8	5.7	17.5	14.1	17.6	8	7.5	7.8	S	DH, PH
Accumulated percentage of variation explained				72.9	40.1	51.7	32.1	52.4	39.9	40.2	35.7	49.9	44.4		

He et al. (2016) QTL Characterization of Fusarium Head Blight Resistance in CIMMYT Bread Wheat Line Soru#1. Plos One 11:e0158052.

# QTL mapping for IAS20\*5/H567.71

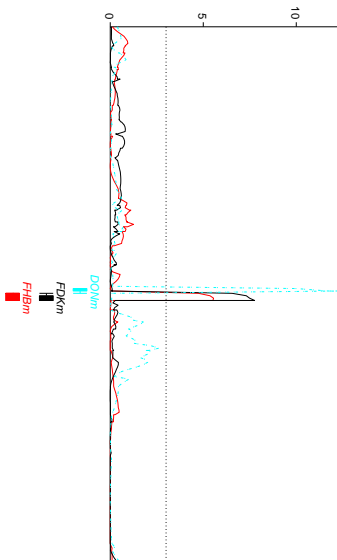
- A RIL population derived from NASMA and IAS20\*5/H567.71 was used, containing 197 entries
- Field trials were performed in 2010, 2013, 2014 and 2017 in El Batan
- Phenotyping was conducted for FHB index, FDK, DON, PH, DH and AE
- Genotyping was done with 15K SNP markers and some SSRs and SNPs used by Dreisigacker et al. (2015)



# QTL mapping for IAS20\*5/H567.71

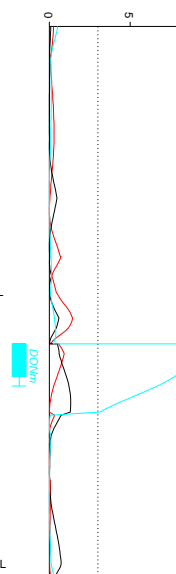
**Chr\_3B**

0 TA004185-0427  
11 Ra\_c76611\_364  
22 BW\_c9711\_71  
29 gwm493  
40 R\_c3591\_1832  
51 R\_c5\_632-1AL  
68 Ra\_c59977\_598  
77 cld6  
85 wmc307  
96 IAAV1382  
100 wmc418  
104 R\_c5871\_1771  
110 BW\_c24364\_73  
111 BS00063034\_51  
112 IACX20464-3DL  
114 BS00048754\_51  
115 WKC\_41142340  
123 wmc291  
130 Ex\_c36725\_96  
141 WEC\_20858600  
151 BS00064896\_51  
162 BS00023017\_51  
173 Ra\_c77832\_143  
181 WJRC\_34666611  
217 Ex\_c20309\_539  
225 BS00063160\_51



**Chr\_3D**

0 TA001877-0445  
8 IAAV2729-3DS  
10 wmc341  
35 Ra\_c33823\_279  
53 gwm645  
71 gwm383  
77 BS00054496\_51-3D  
82 Ra\_c101793\_136-3DL  
90 gwm314  
98 TA003804-0980  
101 GENE-1785\_118  
120 gwm3  
127 wmc552  
138 wmc631  
140 cld009  
166 CAP8\_c1057\_105-3DL  
170 Ra\_c5222\_59-3D



— FHBm  
— FDKm  
— DONm

Chrom.	Position	Left marker	Right marker	FHB index				FDK			DON content			R source	Traits associated
				2010	2013	2014	2017	2013	2014	2017	2013	2014	2017		
1BL	66.3-71.6	wmc419	BS00064032_51									4.4		I	AE
2AL	86.9-98.7	WEC 4379619	BS00039406 51					7.4		5.0				I	
3BL	109.6-115.5	BW_c24364_73	WKC_41142340	8.7	4.8	4.4	6.5	7.5	12.3	13.8	15.8	20	23.4	I	
3DL	89.4-92.5	TA003804-0980	GENE-1785 118								10.5	15.3	9.5	I	
4BS	44.1-46.3	Rht-B1	BS00021984_51		5.5									N	PH, AE
4DS	0-10.7	Rht-D1	barc105	9.5	12.7	9.2	8.3	4.9		5.5				I	PH, AE
5AL	147.6-147.9	WEC_5013188	Vrn-A1	3.9	21.2	21.7								I	DH, PH
5BL	92.1-92.4	BS00022673_51	R_c539_1789		4.8	3.4	5.4							N	DH, AE
7AC	115.6-119.8	WERC 70021470	Ku c97425 164						7.5					N	AE
Accumulated percentage of variation explained				22.1	49.0	38.7	20.2	19.8	19.8	24.3	26.3	39.7	32.9		

## QTL mapping for IAS20\*5/H567.71

- The QTL on 3BL showed consistently higher phenotypic effects on DON than on FHB and FDK
- The QTL on 3DL exhibited only effects on DON, implying an underlying gene for detoxification
- Flanking markers of the 3DL QTL have been transformed into KASP markers, and based on haplotyping results of the two markers on a panel of 574 varieties of global origin, 95 (16.6%) showed resistant alleles at both loci
- Fine mapping of both QTLs are planning





# Conclusion and implications for breeding

- Despite the good Type I resistance, CIMMYT lines need to improve Type II resistance  
The utilization of *Fhb1/Sr2* recombinants will significantly contribute to the improvement of the Type II resistance of CIMMYT germplasm.
- FHB resistance of CIMMYT germplasm is often associated with genes for heading and height, i.e. *Vrn-A1*, *Vrn-D1*, *Rht-B1* and *Rht-D1*  
SNPs for phenology are also important for FHB resistance breeding
- Phenotypic association between AE and FHB is supported by the often coincidence or linkage of QTLs for the two traits
- The 2DLc QTL is very important in the CIMMYT germplasm  
KASP markers are available for this QTL
- QTLs on 3BL and 3DL could be useful in breeding for DON reduction



THANK YOU



QUESTIONS ???