

Detection of a major common bunt resistance QTL on wheat chromosome 6D present in the *Bt11* bunt differential line

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Common Bunt (CB)

Causal agents: *Tilletia caries* and *T. laevis*



Field Trials

- ① winter wheat populations - sowing in **autumn**, first two weeks of November
- ② **artificial inoculation** prior to sowing
- ③ **suspension** of CB teliospores in solution of 2 % **methylcellulose in water**
- ④ spores collected in previous years field trials on **wide range of genotypes**
- ⑤ collection in plots with **medium** (20 % - 50 %) infection levels
- ⑥ thorough **cleaning** from all plant residues
- ⑦ 0.3 mL of inoculum (= 0.09 g of spores) per 10 g of seeds

Artificial Inoculation



Inoculum Sources

IFA Housekeeping

- standard inoculum until 2020
- originally from **3 locations in AT**
- spore morphology: *T. caries*
- **high virulence:** *Bt2* and *Bt7*
- lower virulence: *Bt10*, *Bt13*, *BtP*

IFA Aggressive

- based on **infected 'Tilliko' heads** received in 2019
- collected at a farmers field in Austria
- morphologically same as Housekeeping
- standard inoculum from 2021 onwards
- **high virulence:** *Bt2*, *Bt7*, *Bt10*
- lower virulence: *Bt3* and *Bt13*

Phenotypic Scoring

common bunt incidence (CBI) in 100 randomly chosen spikes per plot



Project Overview

Aim

Genetic mapping of the resistance factor harboured by the differential line for *Bt11*, M822123 (PI 166910 × 'Elgin')

MP-PR1

- PI 166910 × 'Rainer'
- 120 recombinant inbred lines (RILs)
- 3 years

MP-PL:

- PI 166910 × 'Lukullus'
- 160 RILs
- 2 years

MP-PR2

- 'Rainer' × PI 166910
- 120 RILs
- 3 years

MP-MM:

- M822123 × 'Mulan'
- 105 RILs
- 4 years

Genotypic Data Analysis

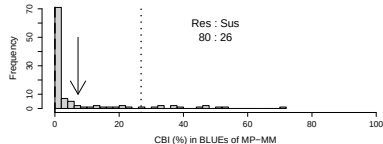
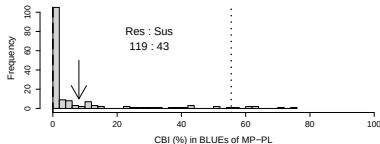
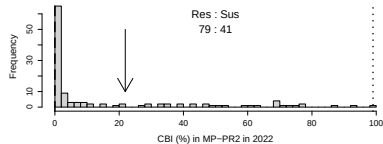
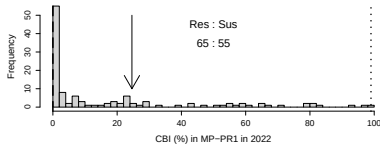
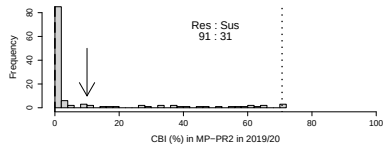
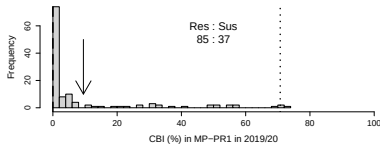
All RILs and parental lines genotyped with 25K wheat SNP array

- data filtered for
 - ① marker call rate ($\geq 80\%$)
 - ② marker segregation distortion ($p \leq 0.001$)
 - ③ genotype call rate ($\geq 80\%$)
- MP-PR1 and MP-PR2 no reciprocal crosses: different **sublines of PI 166910**
- analysis separately for individual MPs (between 1821 and 2795 SNPs)
- linkage groups (LGs) identified with R package *ASMap*
- LGs assigned to chromosomes based on marker positions in IWGSC RefSeq v2.1

Results - General

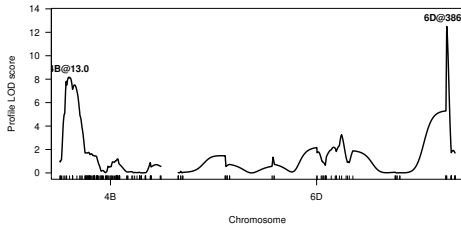
- 2022: **different bunt population** tested (only on MP-PR1 and MP-PR2)
- infection levels approx. 50 % **higher** in 2022
- PI166910 and M822123: **completely resistant**
- susceptible parents **moderately** ('Mulan') to **highly** ('Rainer', 'Lukullus') infected
- **high variation** in CBI (0-99 %)
- **high heritability** estimates for CBI: 0.95 - 0.97
- **negative correlations** for plant height (PH) and CBI: $r = -0.13$ to $r = -0.25$

Results - Data Distribution

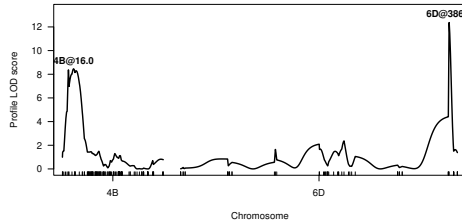


Results - MP-PR1 (PI 166910 × 'Rainer')

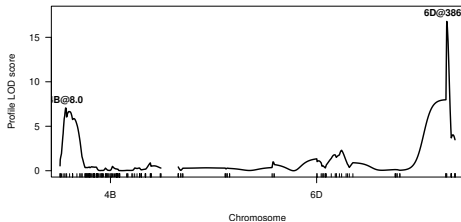
MP-PR1 2019



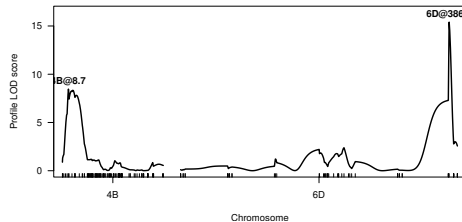
MP-PR1 2020



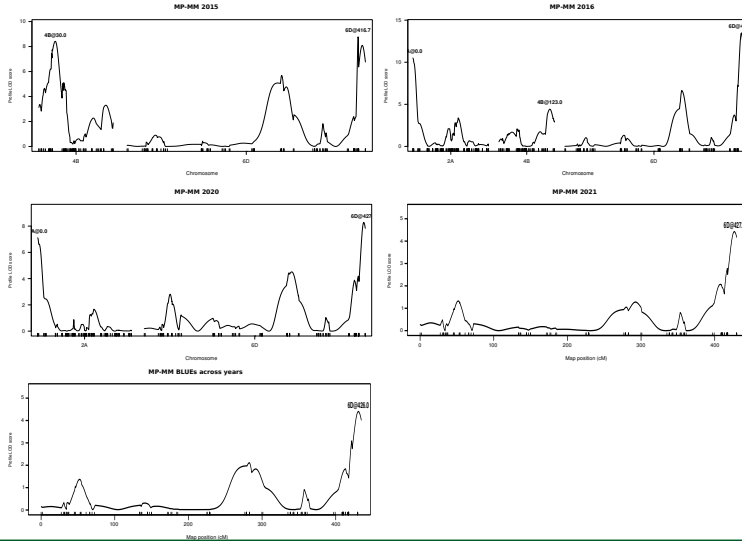
MP-PR1 2022



MP-PR1 BLUEs across years



Results - MP-MM (M822123 × 'Mulan')



Common Bunt Resistance QTL

Chrom	cM ^a (dist.)	Mbp ^b (pos.)	PV % ^c	Experiment
1B	5.4	12.2-46.9	34.8	<u>MP-PR2</u> : all experiments
2A	4.5	0.3-35.1	26.5	<u>MP-MM</u> : 2016, 2020
4B	38.0	11.5-28.6	22.0	<u>MP-PR1</u> : all experiments <u>MP-PL</u> : all experiments <u>MP-MM</u> : 2015, 2016
6D	13.2	482.8-495.2	33.9	all MPs, all experiments
7B	14.45	7.1-26.4	14.2	<u>MP-PR2</u> : 2019, 2020

^a maximum distance spanned by the QTL in cM

^b physical QTL position across experiments in Mbp

^c average amount of phenotypic variation explained by the QTL across experiments

What Did We Map?

Expectation: identify chromosomal location of *Bt11* resistance factor

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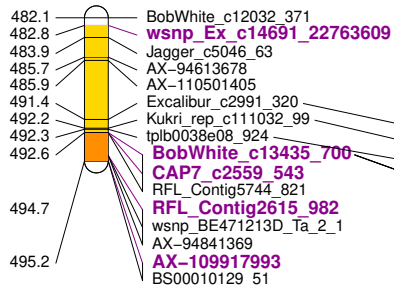
Expectation: identify chromosomal location of *Bt11* resistance factor

Reality: The 6D Locus Is...

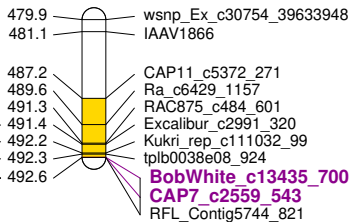
- overlapping with the *Bt9* locus: Steffan et al. (2017) and Wang et al. (2019)

Comparison Of Resistance Loci On Chromosome 6D

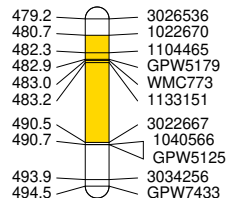
6D-RefSeq_v2.1 (Mbp)



QDB.ui-6DL RefSeq_v2.1 (Mbp)



Bt9-DH RefSeq_v2.1 (Mbp)



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Reality: The 6D Locus Is...

- overlapping with the *Bt9* locus: Steffan et al. (2017) and Wang et al. (2019)
- hard to fine-map because of **marker scarcity** in the region

What Did We Map?

Expectation: identify chromosomal location of *Bt11* resistance factor

Reality: The 6D Locus Is...

- overlapping with the *Bt9* locus: Steffan et al. (2017) and Wang et al. (2019)
- hard to fine-map because of **marker scarcity** in the region
- likely to be **different** from *Bt9* because:
 - ① PI 166910 has the **susceptible allele for *Bt9*-markers** in Wang et al. (2019)
 - ② *Bt9* does not provide **full resistance** against our bunt inoculum
 - ③ both resistance donors for our MPs are **postulated to harbour *Bt11***
 - ④ *Bt11*-genotypes show a **distinct haplotype in the QTL region**, *Bt9*-genotypes do not
- with high probability corresponding to *Bt11*

Haplotype Comparison in the 6DL Region

SNP-ID	QTL-regions	bp, RefSeq v2	F94976GM1-11	PI166910	PI211657	PI554119	PI554099	PI178383	PI542575	702-1102-C	Golden Spike	PI554113
postulated Bt-locus			Bt11	Bt7, 9, 11	Bt11	Bt11-differential	Bt9-differential	Bt8, 9, 10	Bt8, 9	Bt8, 9	Bt9+	Bt9, 10
BS00023009_51			490334091 failed	failed	failed	failed	failed	failed	failed	failed	G	failed
BS00022206_51			490336447 failed	G	R	A	A	A	A	A	A	A
Excalibur_c16566_485			490337615 A	A	A	A	G	G	G	G	G	G
wsnp_JG_c5646_2148296			490706875 A	A	A	A	C	C	C	C	C	C
wsnp_JG_c5646_2148382			490706913 A	A	A	A	C	C	C	C	C	C
Kukri_rep_c107605_164			490707695 C	C	C	C	T	T	T	T	NA	T
wsnp_CAP7_c1735_859875			490708600 A	A	A	A	G	G	G	G	G	G
wsnp_CAP7_c1735_859744			490708656 C	C	C	C	T	T	T	T	T	T
wsnp_CAP8_rep_c4586_223287			490708724 T	T	T	T	C	C	C	C	NA	C
AX-94589700			490830554 C	C	C	C	A	A	A	A	NA	A
RAC875_c484_1063			491341616 G	G	G	G	A	A	A	A	NA	A
Excalibur_c2991_320			491431473 C	C	C	C	C	C	C	C	C	C
Kukri_c45876_61			491554593 T	T	T	T	T	T	T	T	T	T
AX-94886098			491554847 C	C	C	C	C	C	C	C	NA	C
AX-9494854			491955946 A	A	failed	A	C	C	C	A	NA	C
Kukri_rep_c111032_99			492199734 C	C	C	C	C	C	C	C	C	C
BobWhite_c13202_312			492201480 T	T	T	T	T	T	T	T	C	T
Excalibur_c7546_1286			492211389 C	C	C	C	C	C	C	C	C	C
tpib0038e08_609			492253034 A	A	A	A	A	A	A	A	A	A
tpib0038e08_924			492255162 C	C	C	C	C	C	C	C	C	C
wsnp_Ra_c39433_47141896			492255909 T	T	T	T	T	T	T	T	NA	T
TA002907-0764			492545465 C	C	C	C	C	C	C	C	C	C
BobWhite_c13435_700			492576814 C	C	C	C	T	T	T	C	T	T
AX-95152423			492584966 A	A	A	A	A	A	A	A	NA	A
Kukri_c14511_217			492585005 G	G	G	G	A	A	A	A	NA	A
CAP7_c2559_543			492585960 A	A	A	A	C	C	C	A	C	C
Kukri_c44291_138			492637376 G	G	G	G	G	G	G	A	NA	G
RFL_Contig5744_821			492637565 C	C	C	C	C	C	C	T	T	C
Kukri_c373_1588			493720946 G	G	G	G	A	A	A	G	R	A
Kukri_c373_916			493721866 T	T	T	T	C	C	C	T	C	C
AX-109985406			494216977 C	C	C	C	T	T	T	T	NA	T
Kukri_c48283_78			494513676 C	C	C	C	C	C	C	C	C	C
RAC875_rep_c104893_620			494515084 C	C	C	C	C	C	C	C	C	C
AX-109853614			494584093 G	G	G	G	A	A	A	A	NA	A
BS00070856_51			494584603 G	G	G	G	T	T	T	T	T	T
RFL_Contig2615_982			494682864 C	C	C	C	T	T	T	T	T	T
Excalibur_c98257_136			494683154 failed	T	failed	T	T	failed	failed	C	NA	failed
wsnp_BE471213D-Ta_2_1			494683543 C	C	C	C	T	T	T	T	NA	T
AX-94841369			494688665 T	T	T	T	A	A	A	A	NA	A
AX-94747666			494888823 T	T	T	T	T	T	T	T	NA	T
AX-158602745			495061511 C	C	C	C	C	C	C	T	NA	C
AX-109917993			495158557 T	T	failed	T	C	C	C	C	NA	C
BS00010129_51			495206868 T	T	T	T	T	T	T	G	T	T

Conclusions

- ① main resistance factor in our MPs: locus at **distal end of 6DL**
- ② **unambiguous distinction** of 6D locus from *Bt9* not possible in our data
- ③ strong arguments supporting mapping of *Bt11*
- ④ complete and **stable resistance**
- ⑤ valuable addition to known genetic resources for **resistance breeding**
- ⑥ **selection** with markers at 492.6 - 495.2 Mbp
- ⑦ attempts to **pyramid** 6DL locus with other major resistance factors in progress



Supervisors, Collaborators and Funding Agencies

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- AgriGenomics DocSchool & Advisory Board:
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Thank you for your attention!

No more slides

