

Genetic and molecular analysis of Sclerotinia stem rot resistance in *Brassica napus*



Background

- In Western Australia, many resistances have been identified in canola and related Brassicas (Li *et.al.* 2005, 2006, 2008; Garg *et.al.* 2008,2010; Uloth *et.al.* 2013, 2014; You *et.al.* 2016)
- Some resistances (India, China and Australia) particularly valuable as pathotype independent (Barbetti *et.al.* 2014)
- Mechanisms of resistance have been defined histologically (Uloth *et.al.* 2015,2016)
- BUT:
Inheritance of resistance not well defined

Genotypes tested

Three crosses were used:

- i) (*B. napus* NC8 x *B. napus* RQ-001-NCA-8 NC2-7)
- ii) (*B. napus* CRN x *B. napus* RQ-001-NCA-8 NC2-7)
- iii) (*B. napus* CRN x *B. napus* NC4-5)



Stem inoculation



- Screening genotypes under field conditions at the flowering stage
- Single agar plug disc method (Buchwaldt *et al.* 2004, 2005)

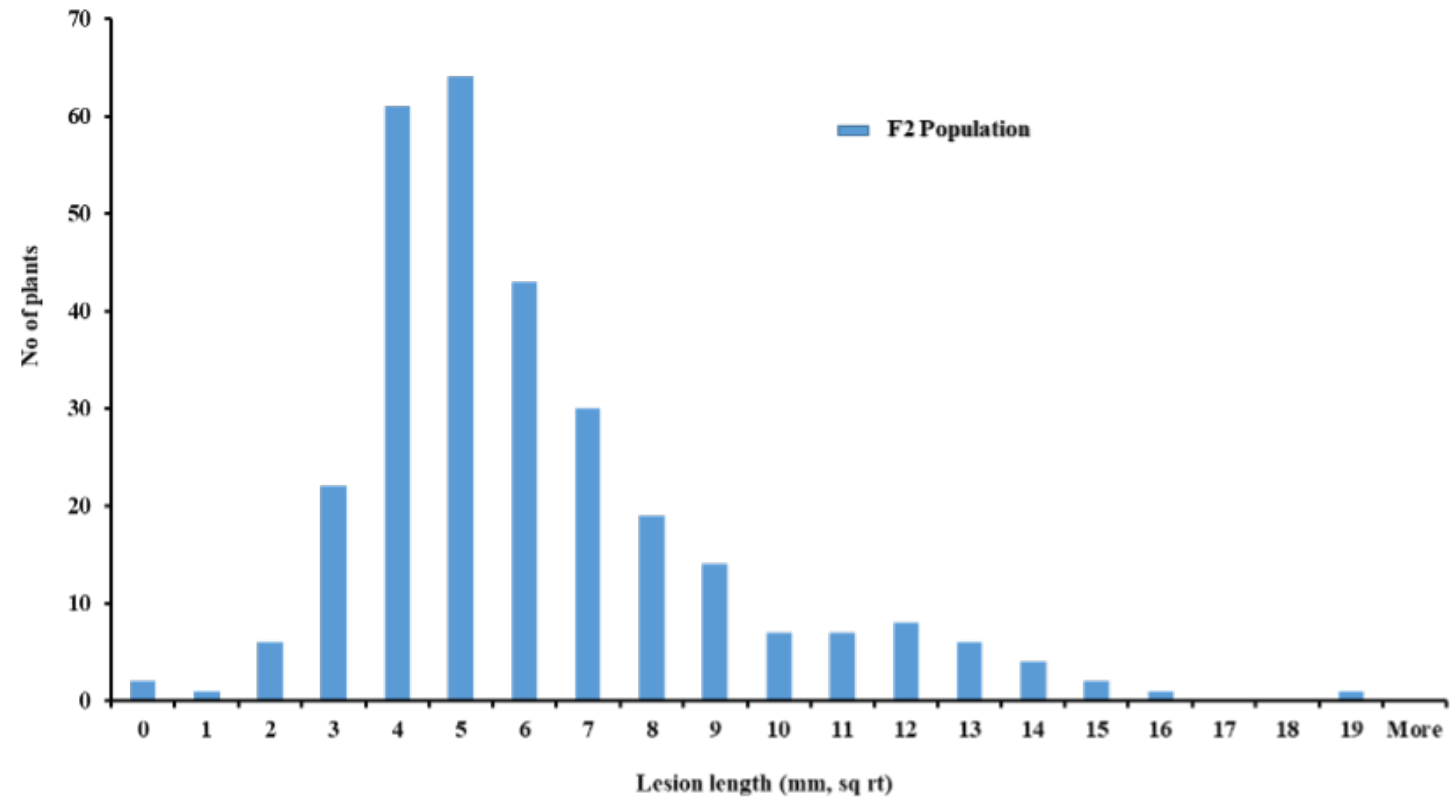
Stem lesions



Phenotypic characteristics

Trait	Attributes	Generation						V_g	V_e	H
		P1	P2	F_1	F_2	BC ₁ P1	BC ₁ P2			
SLL	Mean \pm	3.82 \pm	6.23 \pm	4.52 \pm	5.66 \pm	5.49 \pm	5.83 \pm			
	SE	0.18	0.51	0.24	0.16	0.50	0.52			
	Variance	1.1	6.75	1.73	7.57	8	9.85	3.19	4.38	0.58
	n	34	26	29	298	32	36			
SD	Mean \pm	16.03 \pm	12.19 \pm	11.44 \pm	10.76 \pm	10.76 \pm	11.68 \pm			
	SE	0.43	0.49	0.64	0.15	0.44	0.51			
	Variance	6.38	7.29	12.93	6.35	6.24	9.23	8.87	-2.52	0.00
	n	35	31	32	298	32	36			
DTF	Mean \pm	155.89 \pm	129.19 \pm	127.25 \pm	127.96 \pm	119.34 \pm	126.94 \pm			
	SE	0.89	1.17	2.12	0.65	2.3	2.43			
	Variance	27.46	42.56	143.23	126.66	169.39	213.08	71.08	55.57	0.44
	n	35	31	32	298	32	36			

Stem Lesion Length



Non additive genetic effects

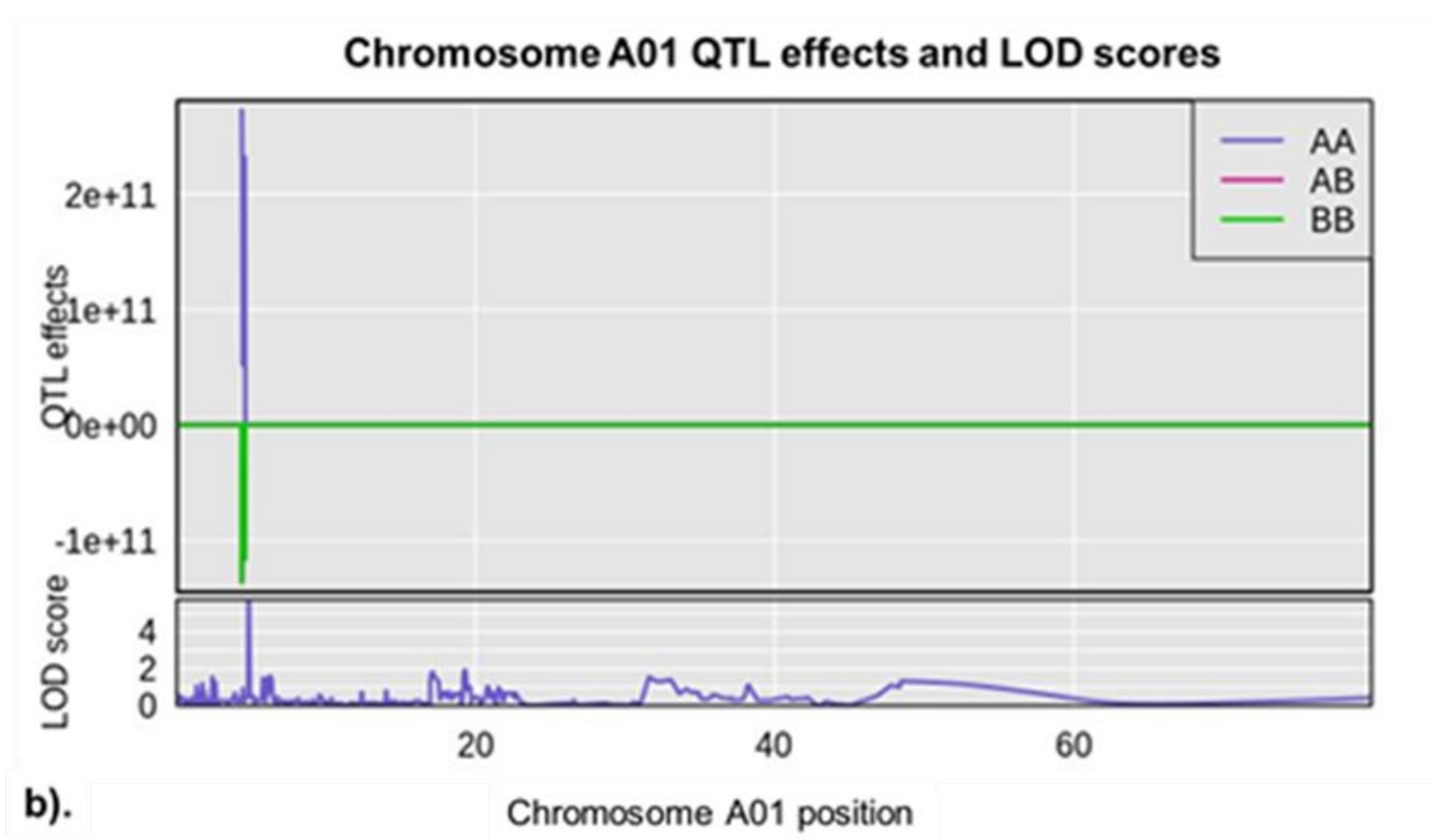
- There was significant negative covariance of non-additive genetic effects between SLL:SD and SLL:DTF
- SLL tended to be larger in genotypes with smaller diameter stems and those that were earlier flowering
- The positive covariance of non-additive genetic effects between SD:DTF indicates that genotypes with wider diameter stems tended to be later flowering

Trait	^a SLL ¹ vs SD ²			^b SLL ¹ vs DTF ²			^c SD ¹ vs DTF ²	
	VC ± SE	Z-ratio	<i>r</i>	VC ± SE	Z-ratio	<i>r</i>	VC ± SE	Z-ratio
	Additive effects			Additive effects			Additive effects	
Trait 1	0.00 ± 0.00	0.00		0.00 ± 0.00	0.00		0.05 ± 0.40 ^{NS}	0.12 ^{NS}
Trait 2	0.07 ± 0.44	0.17 ^{NS}		10.31 ± 16.65	0.62 ^{NS}		9.89 ± 16.81 ^{NS}	0.59 ^{NS}

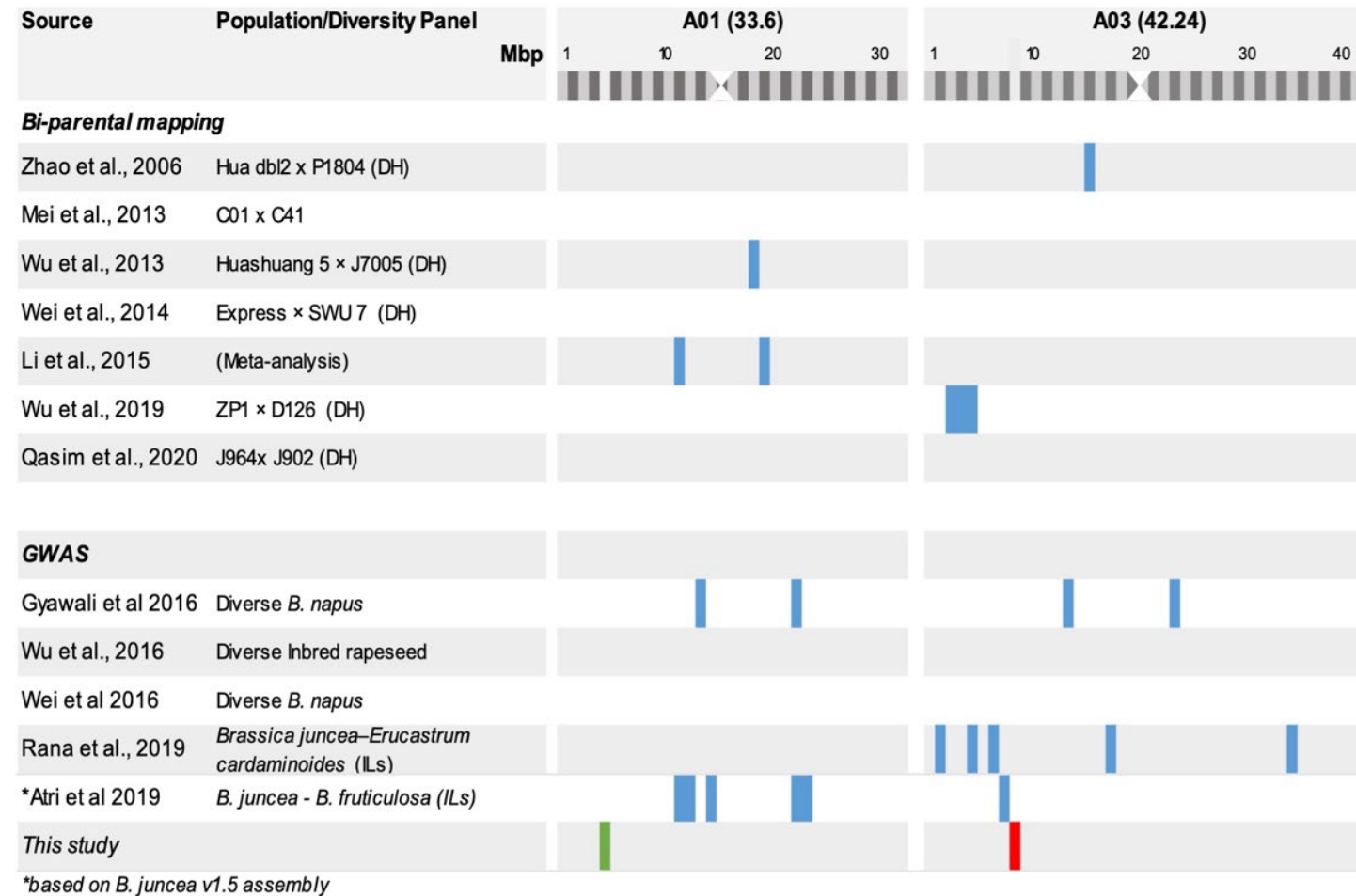
QTL Analysis

- Genotyped using the Brassica 60K Infinium Array
- 36,255 polymorphic SNPs were retained after filtering and used in the downstream analyses.
- The A genome contained a significantly higher number of polymorphic SNPs (22,363) compared to the C genome (10,761). A further 3,101 unplaced SNPs were also included in the analysis.
- On average, the A genome had 2,236 SNPs per chromosome, ranging from 1,765 (A08) to 2,990 (A03), while the C genome contained 1,195 SNPs per chromosome ranging from 424 (C05) to 1,889 (C04).

QTL Analysis



QTL Analysis



Candidate genes

- A03 QTL region contained 41 genes
- Five genes predicted to contain protein kinase domains
- One gene predicted to contain a transmembrane domain
- A01 QTL region contained two putative resistance genes

Summary

- Developed new populations for analysis of Sclerotinia Stem Rot resistance
- Two QTL regions identified for SLL, one overlapping with previous studies
- No associations identified for SD and DTF
- Candidate genes underlying QTL can be further characterised in the population

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