THE UNIVERSITY OF WESTERN AUSTRALIA

THE UNIVERSITY OF QUEENSLAND AUSTRALIA

LepR3 and *Rlm2* alleles

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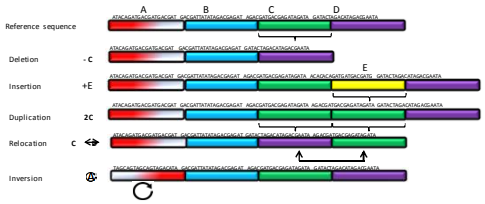
Copy number variation

A form of structural variation resulting in the cell having different number of copies of one or more segments of DNA

- Chromosomal deletions
- Insertions
- Duplications

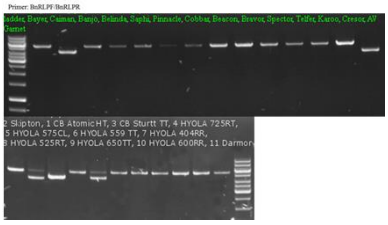


LepR3 and *Rlm2*

- Only cloned blackleg genes
- Allelic variants
  - Alleles are different lengths
- *Rlm2* interacts with *Avr2*
- *LepR3* interacts with *Avr1*?
  - What about *Rlm1*?
  - What alleles are in Australian cultivars?

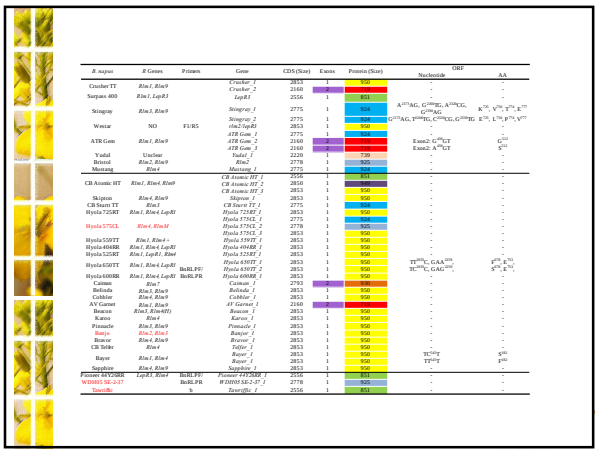
LepR3 and *Rlm2*

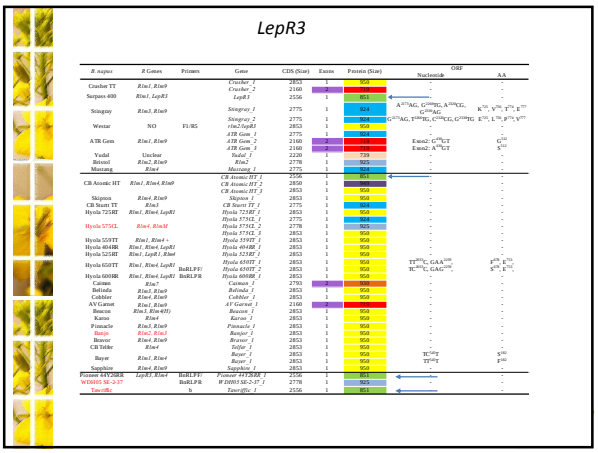
- Designed PCR primers
- Sequenced different cultivars/lines
  - Different Brassica species

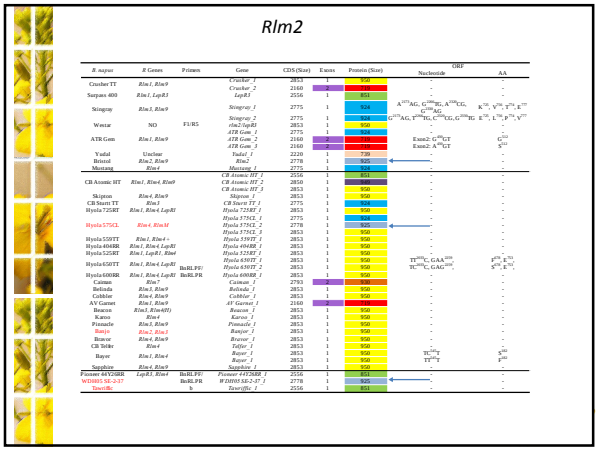


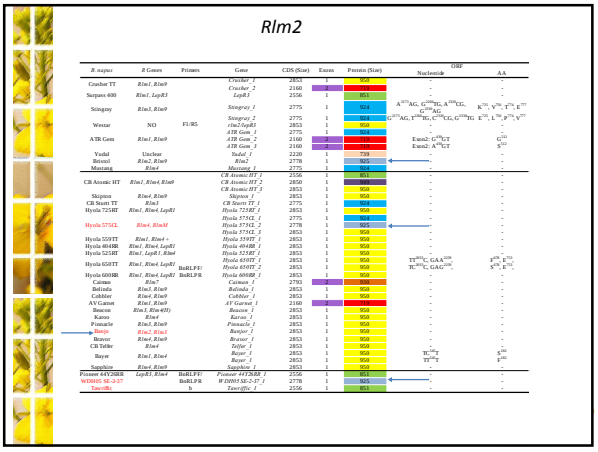
LepR3 and *Rlm2*

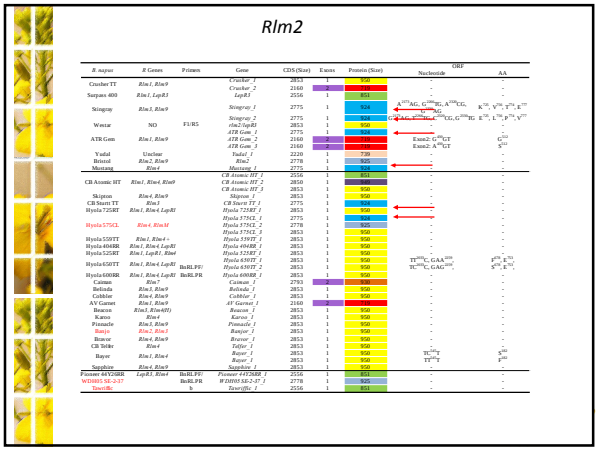
- Nine different size alleles identified
- Sequenced *Rlm2* and *LepR3*
  - Some alleles same size but polymorphisms
  - Some cultivars have more than 1 allele

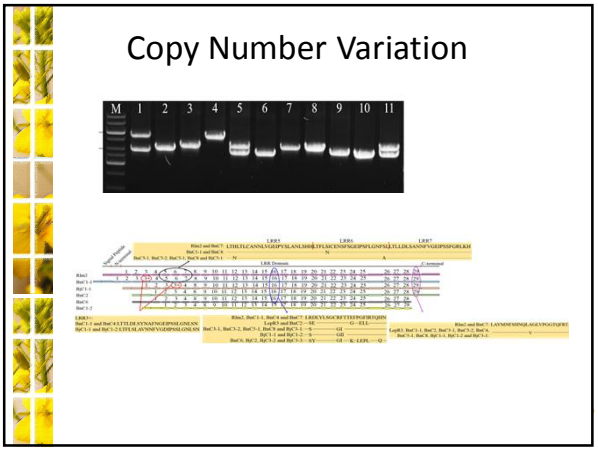



LepR3


Rlm2


Rlm2


Rlm2


Copy Number Variation
M
1
2
3
4
5
6
7
8
9
10
11

Protein (Size)	*B. napus*	Gene	ORF			LRR
			Nucleotide	AA	Locus	
950	Westar *	*rlm2/lepR3*	$TC^{545}T$, $TC^{2033}C$, GAG^{2259}, $G^{2317}GA$	S^{182}, S^{678}, E^{753}, G^{773}	-	30
	Crusher TT	*Crusher_1*	-	-	-	
	CB Atomic HT	*CB Atomic HT_3*	-	-	-	
	Skipton	*Skipton_1*	-	-	-	
	Hyola 725RT	*Hyola 725RT_1*	-	-	-	
	Hyola 575CL	*Hyola 575CL_3*	-	-	-	
	Hyola 559TT	*Hyola 559TT_1*	-	-	-	
	Hyola 404RR	*Hyola 404RR_1*	-	-	-	
	Hyola 525RT	*Hyola 525RT_1*	-	-	-	
	Hyola 650TT	*Hyola 650TT_1*	$TT^{2033}C$, GAA^{2259},	F^{678}, E^{753},	LRR	
		Hyola 650TT_2	-	-	-	
	Hyola 600RR	*Hyola 600RR_1*	-	-	-	
	Belinda	*Belinda_1*	-	-	-	
	Cobbler	*Cobbler_1*	-	-	-	
	Beacon	*Beacon_1*	-	-	-	
	Karoo	*Karoo_1*	$A^{2317}GA$	R^{773}	LRR	
	Pinnacle	*Pinnacle_1*	-	-	-	
	Banjo	*Banjo_1*	-	-	-	
	Bravor	*Bravor_1*	-	-	-	
	CB Telfer	*Telfer_1*	-	-	-	
	Bayer	*Bayer_1*	-	-	-	
		Bayer_1	$TT^{545}T$	F^{182}	LRR	
	Sapphire	*Sapphire_1*	-	-	-	
925	Bristol *	*Rlm2*	C^{553}, T^{688}, G^{732}, C^{771}, T^{773}, G^{785}, G^{787}, A^{789}, T^{808}, G^{819}, A^{834}, $GA^{844\text{-}845}$, A^{852}, T^{855}, G^{857}, T^{859}, G^{892}, T^{899}, $GC^{916\text{-}917}$, C^{923}, G^{928}, G^{933}, A^{942}, G^{2352}, C^{2382}, G^{2385}, C^{2390}, A^{2397}, C^{2456}, G^{2463}, A^{2466}, A^{2487}, A^{2528}, G^{2538}, $GA^{2542\text{-}2544}$, G^{2545}, T^{2549}, T^{2590}, G^{2595}, A^{2653}, T^{2672}, T^{2682}, G^{2690}, C^{2692}, G^{2698}, C^{2716}, C^{2732}, G^{2778},	H^{185}, S^{230}, I^{258}, G^{262}, F^{270}, D^{282}, G^{286}, Y^{287}, V^{298}, L^{300}, A^{306}, S^{308}, G^{310}, L^{314}, A^{797}, A^{819}, Y^{843}, E^{848}, E^{849}, V^{850}, L^{864}, S^{885}, V^{891}, G^{897}, V^{900}, Q^{906}, P^{911}	-	29
	Hyola 575CL	*Hyola 575CL_2*	A^{553}, G^{688}, T^{732}, T^{771}, A^{773}, A^{785}, C^{787}, G^{789}, A^{808}, A^{819}, G^{834}, $TC^{844\text{-}845}$, G^{852}, C^{855}, A^{857}, G^{859}, A^{892}, C^{899}, $TT^{916\text{-}917}$, A^{923}, C^{928}, C^{933}, C^{942}, C^{2352}, T^{2382}, A^{2385}, A^{2390}, G^{2397}, T^{2456}, A^{2463}, G^{2466}, G^{2487}, T^{2528}, A^{2538}, $AT^{2542\text{-}2544}$, C^{2545}, C^{2549}, A^{2590}, A^{2595}, G^{2653}, C^{2672}, C^{2682}, A^{2690}, T^{2692}, A^{2698}, G^{2716}, T^{2732}, A^{2778},	N^{185}, A^{230}, D^{258}, E^{262}, I^{270}, S^{282}, D^{286}, D^{287}, I^{298}, S^{300}, F^{306}, Y^{308}, R^{310}, F^{314}, E^{797}, V^{819}, F^{843}, N^{848}, Q^{849}, A^{850}, M^{864}, G^{885}, A^{891}, E^{897}, M^{900}, E^{906}, L^{911}	185-819 LRR, 843-911 D, E, F, G	
	WDH05 SE-2-37	*WDH05 SE-2-37_1*	-	-	-	
924	ATR Gem *	*ATR Gem_1*	$G^{2173}AG$, $T^{2266}TG$, $C^{2320}CG$, $G^{2330}TG$	E^{725}, L^{756}, P^{774}, V^{777}	-	29
	Stingray	*Stingray_1*	$A^{2173}AG$, $G^{2266}TG$, $A^{2320}CG$, $G^{2330}AG$	K^{725}, V^{756}, T^{774}, E^{777}	C2, LRR, LRR, LRR	
		Stingray_2	-	-	-	
	Mustang	*Mustang_1*	-	-	-	
	CB Sturtt TT	*CB Sturtt TT_1*	-	-	-	
	Hyola 575CL	*Hyola 575CL_1*	-	-	-	
851	Surpass 400 *	*LepR3*	-	-	-	26
	CB Atomic HT	*CB Atomic HT_1*	-	-	-	
	Pioneer 44Y26RR	*Pioneer 44Y26RR_1*	-	-	-	
	Tawriffic	*Tawriffic_1*	-	-	-	
719 (Two exons)	Crusher TT *	*Crusher_2*	Exon2: $G^{430}GT$	G^{512}	-	24
	ATR Gem	*ATR Gem_2*	-	-	-	
		ATR Gem_3	Exon2: $A^{430}GT$	S^{512}	LRR	
	AV Garnet	*AV Garnet_1*	-	-	-	
739	Yudal	*Yudal_1*	NA	NA	NA	25
949	CB Atomic HT	*CB Atomic HT_2*	NA	NA	NA	30
930 (Two exons)	Caiman	*Caiman_1*	NA	NA	NA	29

Summary

- Known alleles and novel alleles identified
- Some findings do not fit with phenotypic predictions
- Markers developed to characterise alleles in all germplasm
- What is role of multiple copies?
- Further characterisation of *LepR3*/*Rlm1*