

Part **1**: Progress on Rust Resistance
Breeding of Confection Sunflowers
and
Part **2**: Sunflower SNP Genetic Mapping

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Part 1: Progress on Rust Resistance Breeding of Confection Sunflowers

Objectives

1. Transfer rust resistance genes from oil sunflower to confection-type
 - Limited resistance in commercial confection hybrids
 - Confections are generally more susceptible to diseases
2. Molecularly mark the resistance genes

Rust resistance lines used in this project

Lines	Usage	Rust resistance		Origin of rust resistance
		Race 336	Race 777	
MC 29	R-gene donors	R	S	Wild <i>H. annuus</i> in Texas
HA-R2		R	S	Wild <i>H. argophyllus</i>
HA-R3		R	MR	Wild <i>H. annuus</i> , <i>H. argophyllus</i> and <i>H. petiolaris</i>
HA-R6	R-gene mapping	R	R	A breeding line from France
HA-R8		R	R	A landrace in Arizona, USA
RHA 397		R	R	South Africa
RHA 464		R	R	Wild <i>H. annuus</i> collected in California

Progress on the introgression of rust resistance genes to confectionary sunflower

Recurrent parent (RP) × R-gene donor

Recurrent parents(RP)

- CONFSCCL B1
- CONFSCCL R5
- Highly susceptible to race 336 and race 777

R-gene donors

MC 29, HA-R2, and HA-R3

CONFSCCL B1 as the recurrent parent of MC 29 and HA-R2

CONFSCCL R5 as the recurrent parent of HA-R3

RP × F₁

BC₄F₁

BC₄F₂

BC₄F₃

BC₄F₄

97% RP genome

MAS of homozygous resistant plants

Obtain homozygous resistant F₃ families

Planting F₃ seeds in the field

Summer 2011

In the end of seed ripening, Jan, 2012

Spring 2012

Field performance test, Summer 2012

Release new rust resistant confection lines

2013

Molecular mapping of new rust resistance genes

Evaluation of rust resistance in F₂ and F_{2:3} families

F ₂ Populations	Harvest of F ₂ seeds	No. of F ₂ plants	Harvest of F ₃ seeds	Rust test for F _{2:3} families
HA 89 × RHA 464	Spring, 2010	141	Fall, 2010	141 × 20
HA 89 × HA-R6	Fall, 2010	142	Spring/Summer2011	70 × 20
HA 89 × RHA 397	Fall, 2010	142	Spring/Summer2011	70 × 20
HA 89 × HA-R8	Fall, 2010	142	Spring/Summer2011	Will start in Feb

20 seedlings was rust inoculated in each F₃ family

Mapping of rust resistance gene in RHA 464

Polymorphism screening

- 870 mapped SSR markers
- 398 showed polymorphism

Bulked segregant analysis

- R-bulk: 10 homozygous resistant F₂ plants
- S-bulk: 10 homozygous susceptible F₂ plants

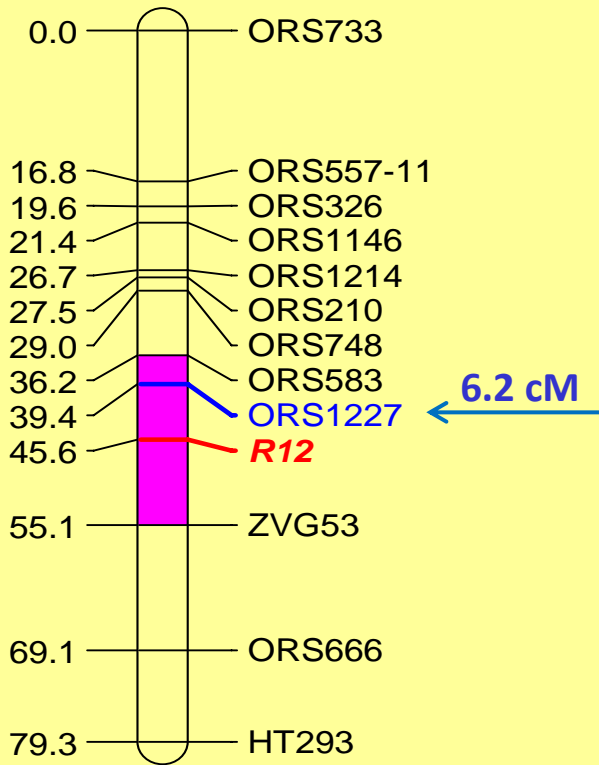
- Designated as *R₁₂*
- 20 markers previously mapped in LG11 showed polymorphism
- 7 polymorphic between R and S-bulk
- Genotyped in 141 F₂ individuals of HA 89 and RHA 464



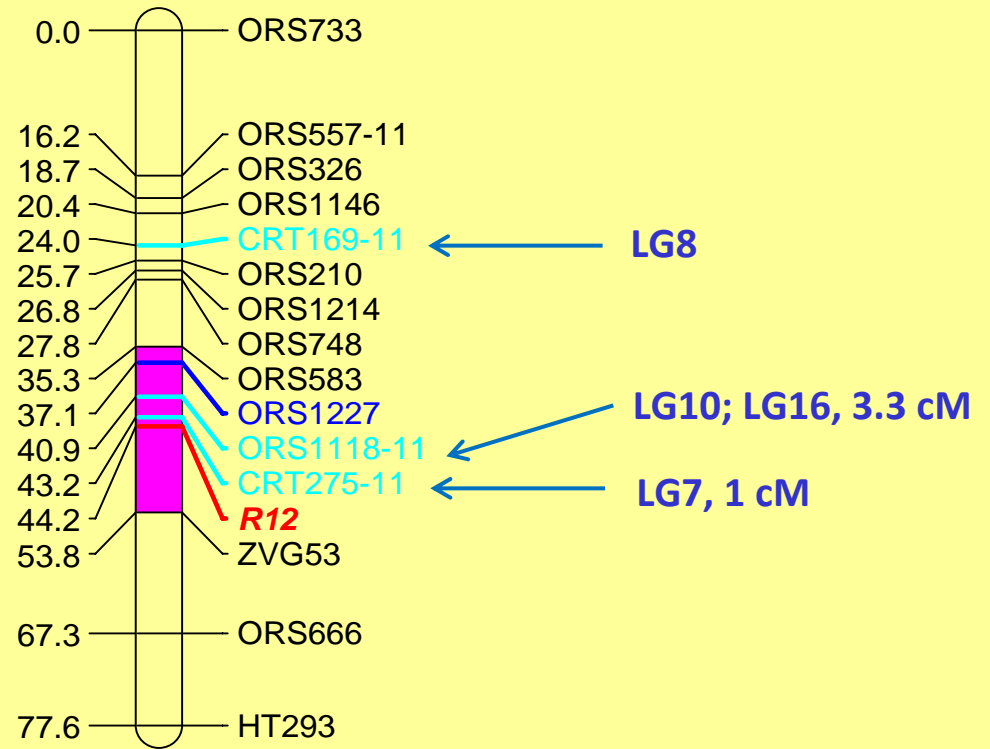
1 RHA 464
2 HA 89
3 R-bulk
4 S-bulk

Mapping R_{12} using SSR markers

LG11



LG11



- After BSA, genotyping only focus on those markers mapped in LG11 in the published maps
- 12 SSR markers associated with R_{12} were mapped in this population

- In the SNP mapping project, all polymorphic SSR markers were genotyped, then CRT169-11, ORS1118-11 and CRT275-11 previously mapped in other LGs were mapped in LG11
- 15 SSR markers associated with R_{12} were mapped

Allelic analysis of rust resistance genes in HA-R6, HA-R8, RHA 397 and RHA 464

HA-R6 × HA-R8



HA-R6 × RHA 397



HA-R6 × RHA 464



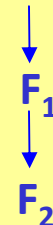
RHA 397 × RHA 464



HA-R8 × RHA 397



HA-R8 × RHA 464



200 plants / F₂ population evaluated for rust resistance

Part 2: Sunflower SNP Genetic Mapping

Mapping population

- F₂ from HA 89 and RHA 464
- HA 89 is susceptible to rust and downy mildew.
- RHA 464 is resistant to rust and downy mildew.
- 141 individuals

Available markers

- **SNP**
2797 → 2446
- **SSR, as bridge/anchor markers**
291 → 238
- **Genes: R_{12} and Pl_{ARG}**

Mapping construction

- **JoinMap 4.1**
- **Mapping algorithm: Regression Mapping**
- **Mapping function: Kosambi**
- **Mapping parameters**

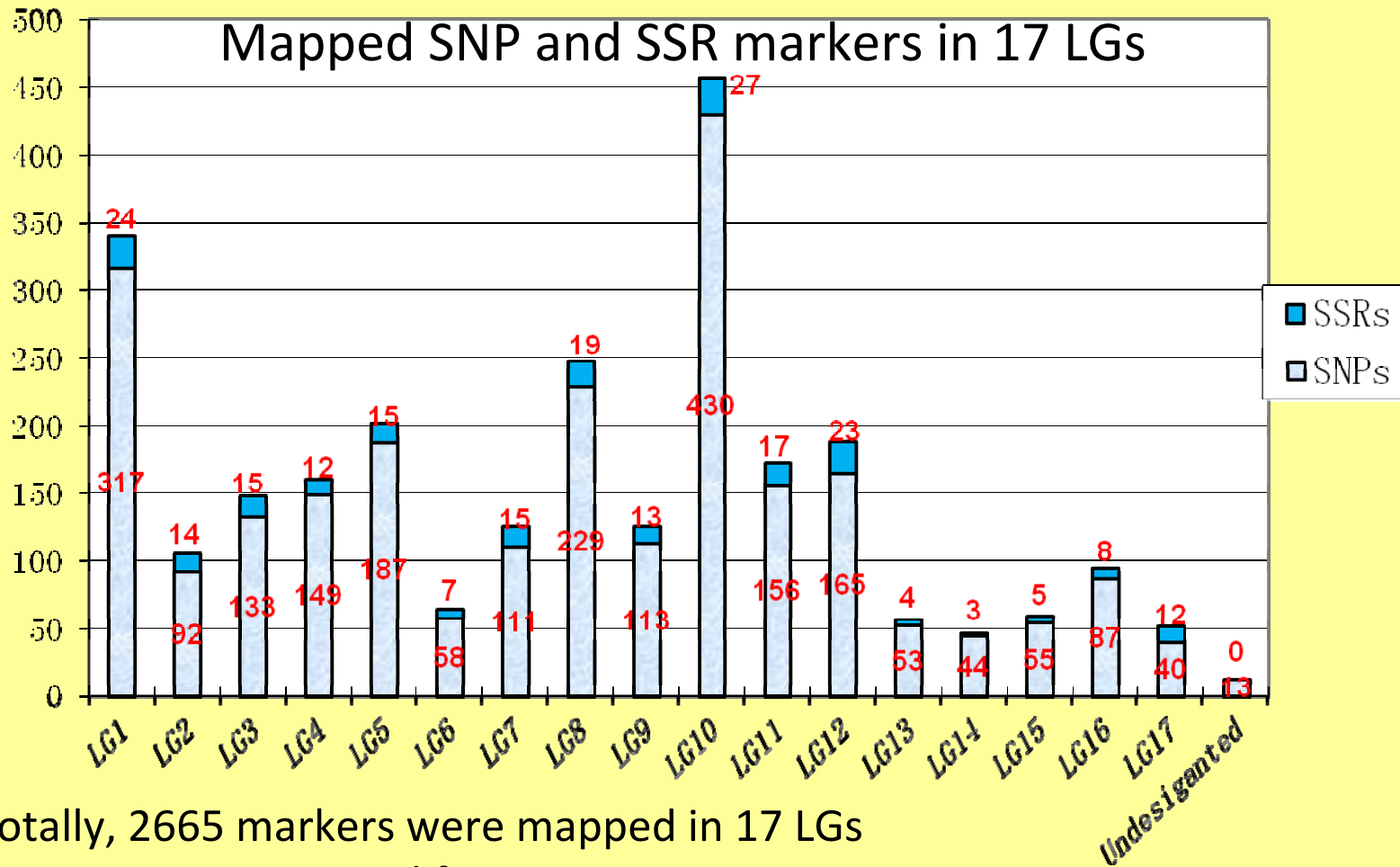
Lod threshold: 1.00

Rec threshold: 0.4000

Jump threshold: 5.000

Ripple value: 1

SNP mapping results of HA89× RHA464

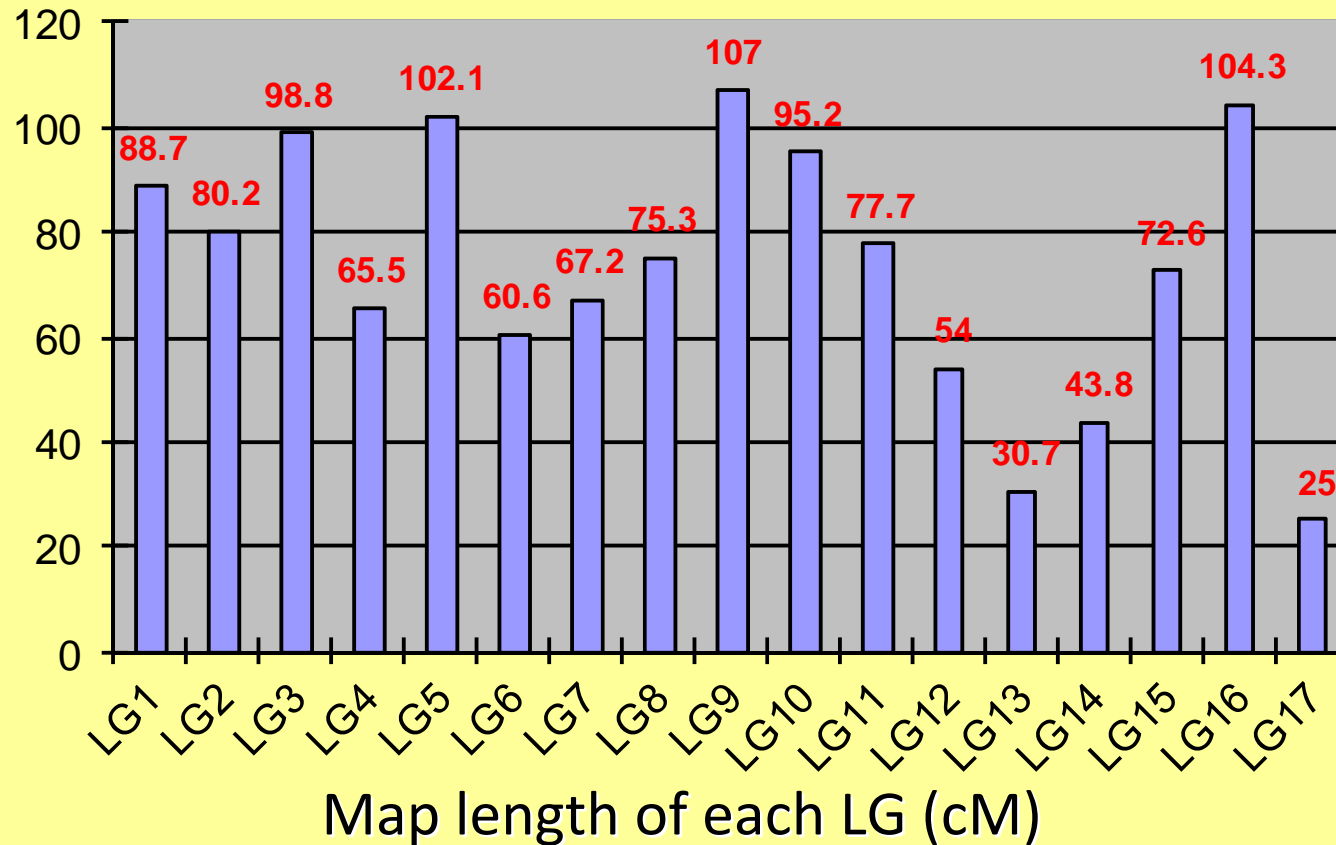


Totally, 2665 markers were mapped in 17 LGs

- SNP 2431, ranged from 40 in LG17 to 430 in LG10
- SSR 233, ranged from 3 in LG13 to 27 in LG10
- R_{12}
- LG1, LG8 and LG10 with a larger amount of markers

SNP mapping results of HA89× RHA464

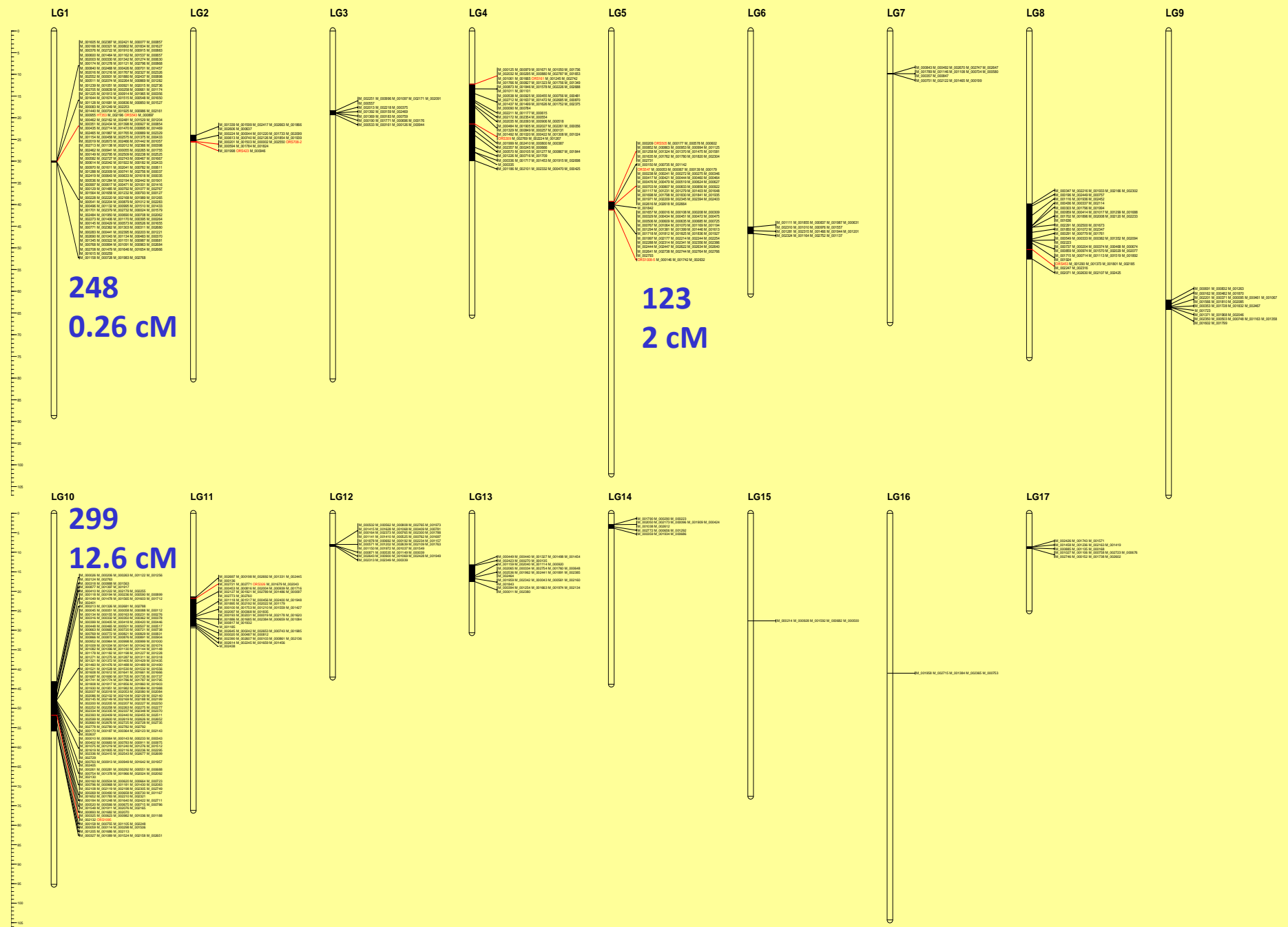
■ Length (cM)



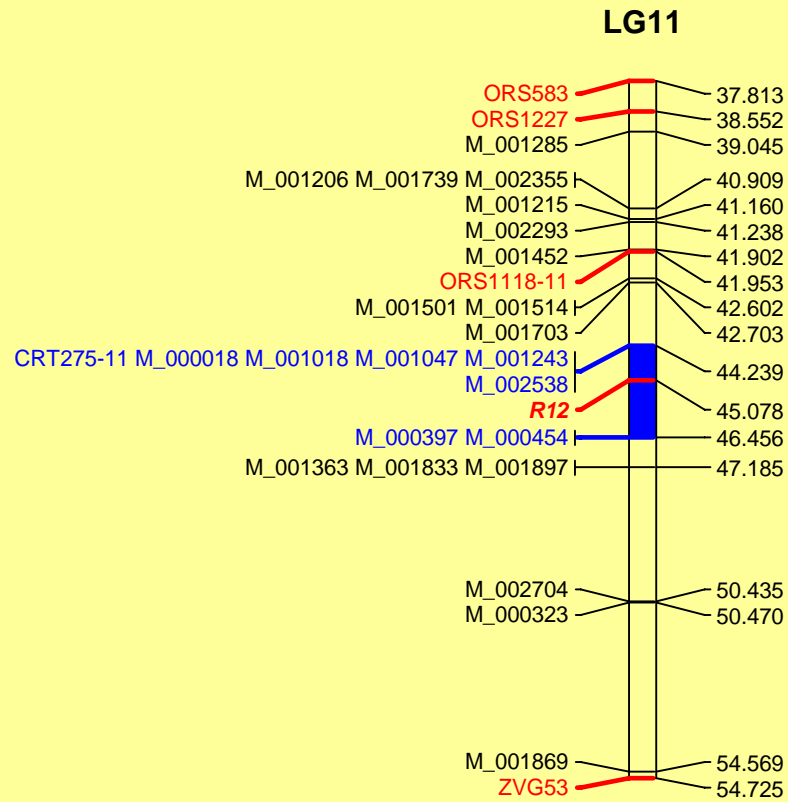
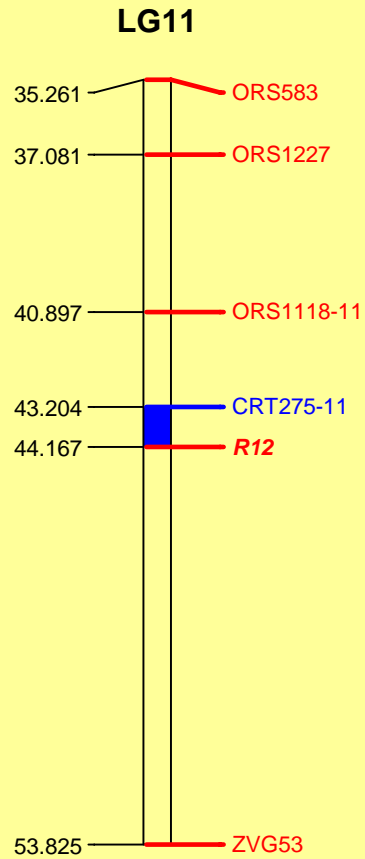
Totally, this map covered 1251.3 cM

- Ranged from 25 cM in LG17 to 107 cM in LG9
- LG13, LG14 and LG17 with a shorter map coverage

Co-segregated SNP markers clustered and distributed in all 17 LGs



SNP markers linked to rust resistance gene R_{12}



Summary of SNP mapping results

- 2665 markers located in ~1250 loci
- composed of 2431 SNP, 233 SSR and R_{12}
- 1251.3 cM,
- ~1 cM per locus
- R_{12} was flanked by two groups of co-segregated markers

Special for this population

- Lower map coverage of LG13, LG14, and LG17
- Co-segregated SNP markers, totally 1415 SNPs (58.3%), clustered and distributed in all LGs

0.9cM

CRT275-11
M_000018
M_001018
M_001047
M_001243
M_002538

1.4cm

M_000397
M_000454

Future work

- MAS of homozygous BC₄F₂ individuals, obtain rust resistant homozygous BC₄F₃ families before growing season this year. The confection breeding lines resistant to the predominant race will be released in 2013
- Continue identifying molecular markers linked to new rust resistance genes in HA-R6, HA-R8 and RHA 397
- Continue investigating the allelic relationships among new rust resistance genes in this project.
- Finalize the SNP mapping in F₂ population of HA 89 and RHA 464
- Map integration of all mapped rust genes in this project

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