

# Molecular mapping of the rust resistance genes in sunflower: results and prospects

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# Sunflower production in North America has recently been threatened by the evolution of new virulent pathotypes of sunflower rust

- ◆ Thirty-nine different rust races were identified in 2008
- ◆ Rust races 334 and 336 were dominant over three years
- ◆ Race 777 is the most virulent and is able to infect all nine differentials
- ◆ Presently, race 777 remains at a low frequency in the rust populations, but can potentially cause serious epidemics if it becomes prevalent
- ◆ The majority of commercial hybrids are susceptible to the new predominant and virulent races



# Rust resistance sources from USDA Sunflower unit

Sunflower line	Type	Reaction to		Putative rust gene (s)
		race 336	race 777	
<b>Inbred lines</b>				
CM 29*	OB	R	S	$R_2$
HA-R2	OB	R	S	$R_5$
HA-R3	OB	R	MR	$R_4$
HA-R6	CB	R	R	
HA-R8	OR	R	R	
RHA 397	OR	R	R	
RHA 464	OR	R	R	
<b>Introgression lines</b>				
PH3	OB	R	R	
PH4	OB	R	R	
PH5	OB	R	R	
PH6	OB	R	S	
TX16R	OB	R	R	
Rf ANN-1742	OR	R	R	

OB: oil-B line; OR: oil-R line; CB: confection-B line;  
 CR: confection-R line. \* an old Canadian line



## Limited durability of single *R*-genes

- ◆ Race-specific *R*-genes can be overcome by new pathotypes
- ◆ There is an urgent need for strategies to develop inbred lines with durable resistance to the disease
- ◆ The concept of incorporating multiple *R*-genes into a single cultivar to achieve greater durability is referred to as '*gene pyramiding*'
- ◆ The selection of genotypes with gene combinations can be difficult using conventional methods
- ◆ Mapping rust resistance genes and developing robust molecular markers will facilitate this breeding approach and add precision to selection



# Procedure for mapping of rust *R*-genes

## I. Mapping population development

Parent 1 (Susc) × Parent 2 (Resist)



F<sub>1</sub>



F<sub>2</sub> individuals



F<sub>3</sub> families



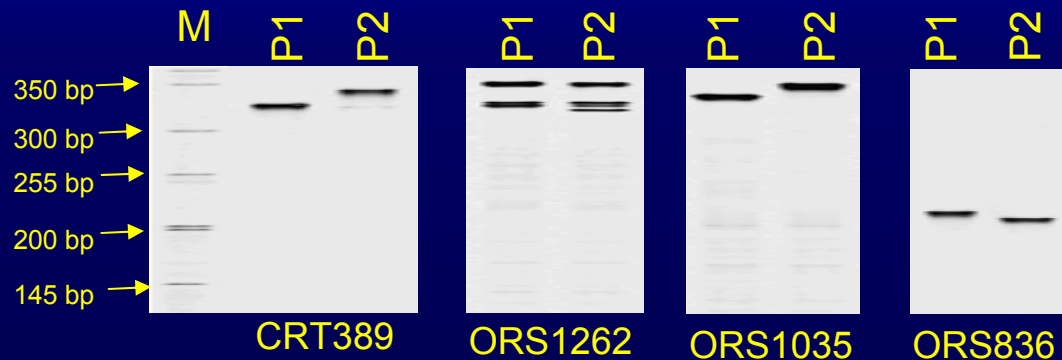
## II. Rust screening of $F_2$ and $F_3$ populations

- I.  $F_2$  population: 150-200 plants
- II.  $F_2$ -derived  $F_3$  population: 150-200 families  
20 plants/per family, a total of 3,000-4,000 plants



### III. Marker screening (500-800 DNA markers)

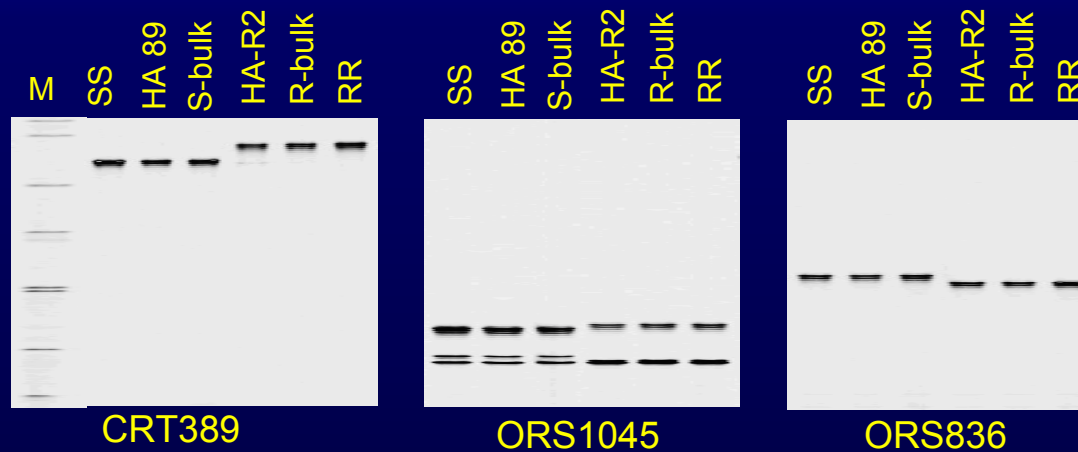
- a. Identification of polymorphic markers for the two parental lines



## b. Bulked segregant analysis to determine genomic region associated with the rust resistance gene

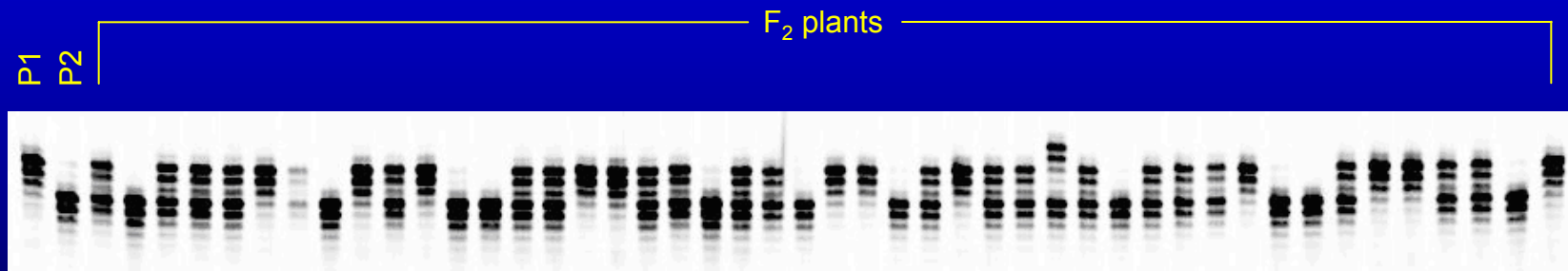
S-bulk: DNA from ten homozygous susceptible  $F_2$  plants

R-bulk: DNA from ten homozygous resistant  $F_2$  plants





c. Scoring all individuals of a mapping population with polymorphic markers associated with rust resistance to determine their genotypes



## IV. Genetic linkage analysis

Combine rust phenotypic and marker data to determine marker order and map distance



# Progress of molecular mapping of the rust *R*-genes in sunflower

- ◆ Eight rust resistance genes,  $R_1$ - $R_5$ ,  $R_{10}$ ,  $P_{u6}$ , and  $R_{adv}$ , were previously identified in sunflower
- ◆  $R_1$  gene in MC 69 and  $R_{adv}$  gene in P2 and RHA 340 lines were mapped to linkage groups (LGs) 8 and 13, respectively, and are no longer effective against the predominant and virulent rust races in U.S.
- ◆  $R_2$  gene in MC 29 was mapped to LG9 and is resistant to race 336

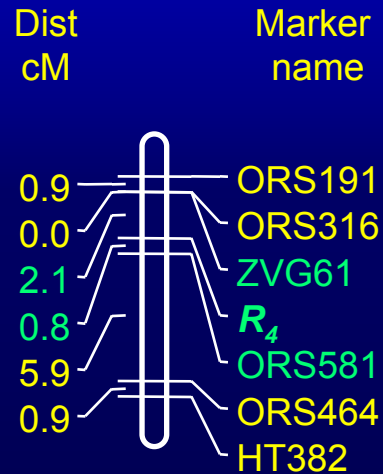
Lawson et al. 1998, Mol Breeding  
Lawson et al. 2010, Mol Breeding  
Yu et al. 2003, Crop Science



# We have recently mapped four rust resistance genes in the different linkage groups of sunflower

**$R_4$  in HA-R3**  
**resistant to race 336**

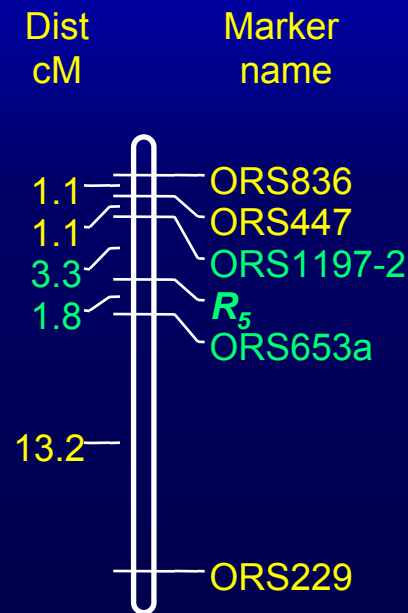
**LG13**



Qi et al. 2011  
 TAG 123:351-358

**$R_5$  in HA-R2**  
**resistant to race 336**

**LG2**

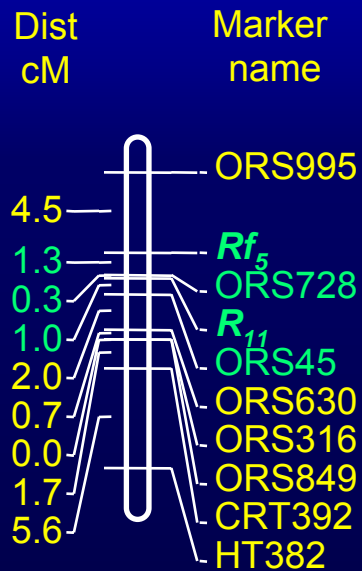


Qi et al. 2011  
 Mol. Breeding DOI  
 10.1007



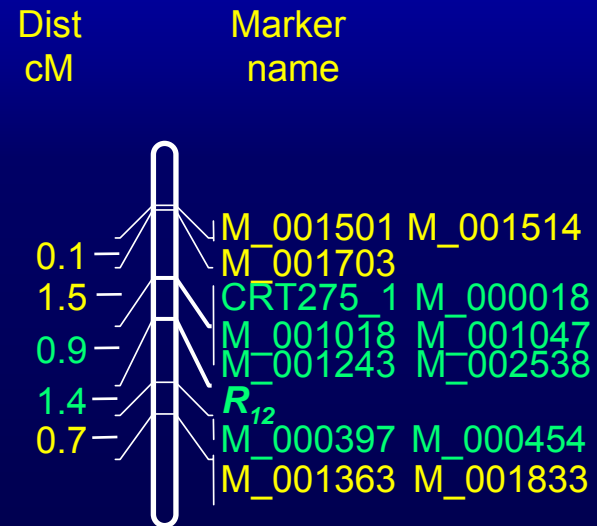
$R_{11}$  in Rf ANN-1742  
resistant to races 336  
and 777

**LG13**



$R_{12}$  in RHA 464  
resistant to races 336  
and 777

**LG11**



# Future work

- I. Integrate phenotypic and SSR data of the genes  $R_4$ ,  $R_5$ , and  $R_{11}$  with SNP data to identify SNP markers linked to the  $R$ -genes
  
- II. Continue mapping of the rust resistance genes in HA-R6, HA-R8, and RHA397
  - ◆ HA-R6 is the only confection sunflower resistant to races 336 and 777, and the  $R$ -gene originated from a breeding line from France
  - ◆ Rust resistance in HA-R8 is derived from a Native American (Havasupai) landrace from Arizona
  - ◆ Rust resistance in RHA 397 originates from germplasm obtained from South Africa



### III. Provide long-term durability of resistance to rust

#### a. Rust *R*-gene pyramiding in oil sunflower

HA-R2 × RHA464    HA-R3 × RHA464    HA-R3 × HA-R2  
( $R_5+R_{12}$ )            ( $R_4+R_{12}$ )            ( $R_4+R_5$ )

#### b. Rust *R*-gene pyramiding in confection sunflower

HA-R6  
(new *R*-gene) × New confection line 1 ( $R_2$ )  
New confection line 2 ( $R_4$ )  
New confection line 3 ( $R_5$ )



# A sample of gene pyramiding with marker-assisted selection

HA-R3 × RHA 464  
( $R_4R_4r_{12}r_{12}$ ) ( $r_4r_4R_{12}R_{12}$ )

↓  
F<sub>1</sub>

( $R_4r_4R_{12}r_{12}$ )

↓  
F<sub>2</sub>

Segregating

↓ Screen a large population  
with DNA markers

Select homozygous F<sub>3</sub>

( $R_4R_4R_{12}R_{12}$ )



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