



New resistance genes against potato late blight

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Solanum michoacanum (Bitter.) Rydb. 2x (1EBN) **VIR5763**

CORRELL: THE POTATO AND ITS WILD RELATIVES

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FIGURE 108.—*Solanum* \times *michoacanum* (Arsène 2896, type collection): 1, flowering plant, $\times \frac{1}{2}$; 2, calyx, spread out, $\times 2$; 3, corolla, spread out, $\times 2$; 4, stamen, dorsal view, $\times 2$; 5, pistil, $\times 2$.

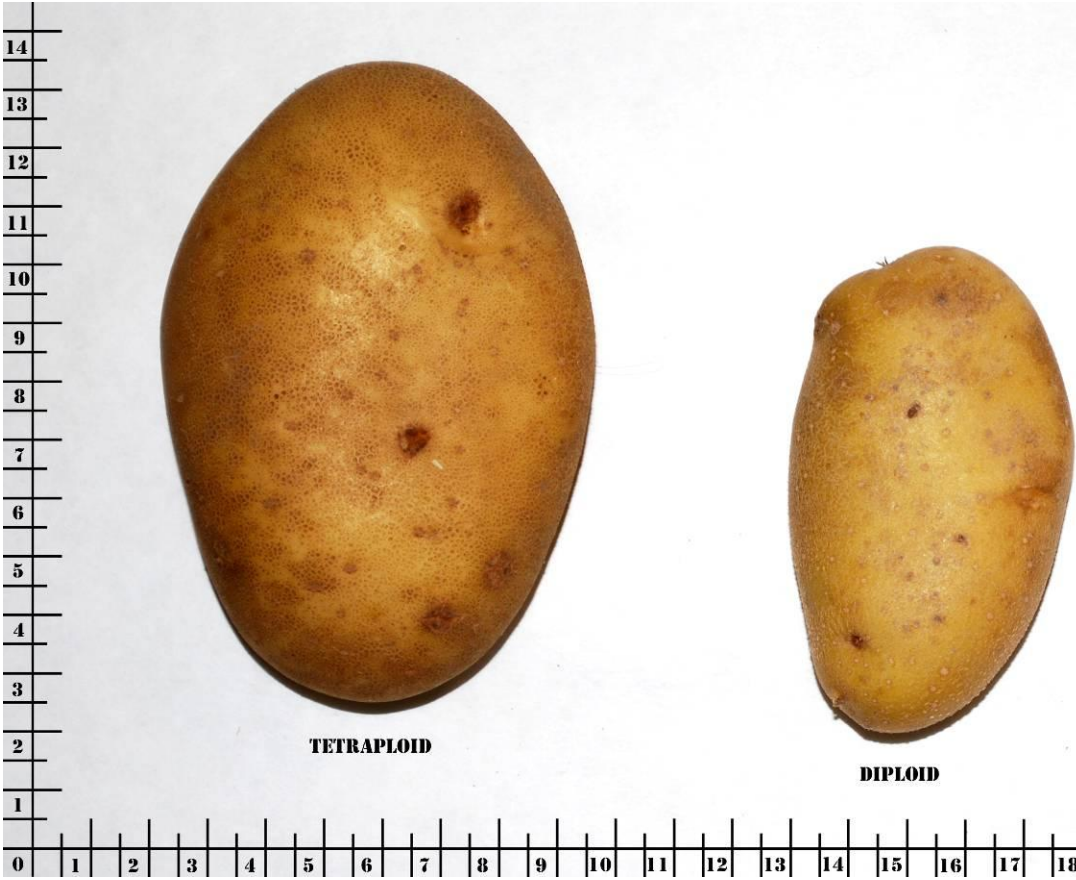
- **Originates from:**
Mexico, state: Michoacán,
habitat: humid, grassy, among rocks,
2000-2100 m above sea level

- **Natural hybrid:**
S. bulbocastanum \times *S. pinnatisectum*
(Hawkes 1990)

- **Selected clones:**
 - resistant to *P. infestans*
 - suitable for cold chipping

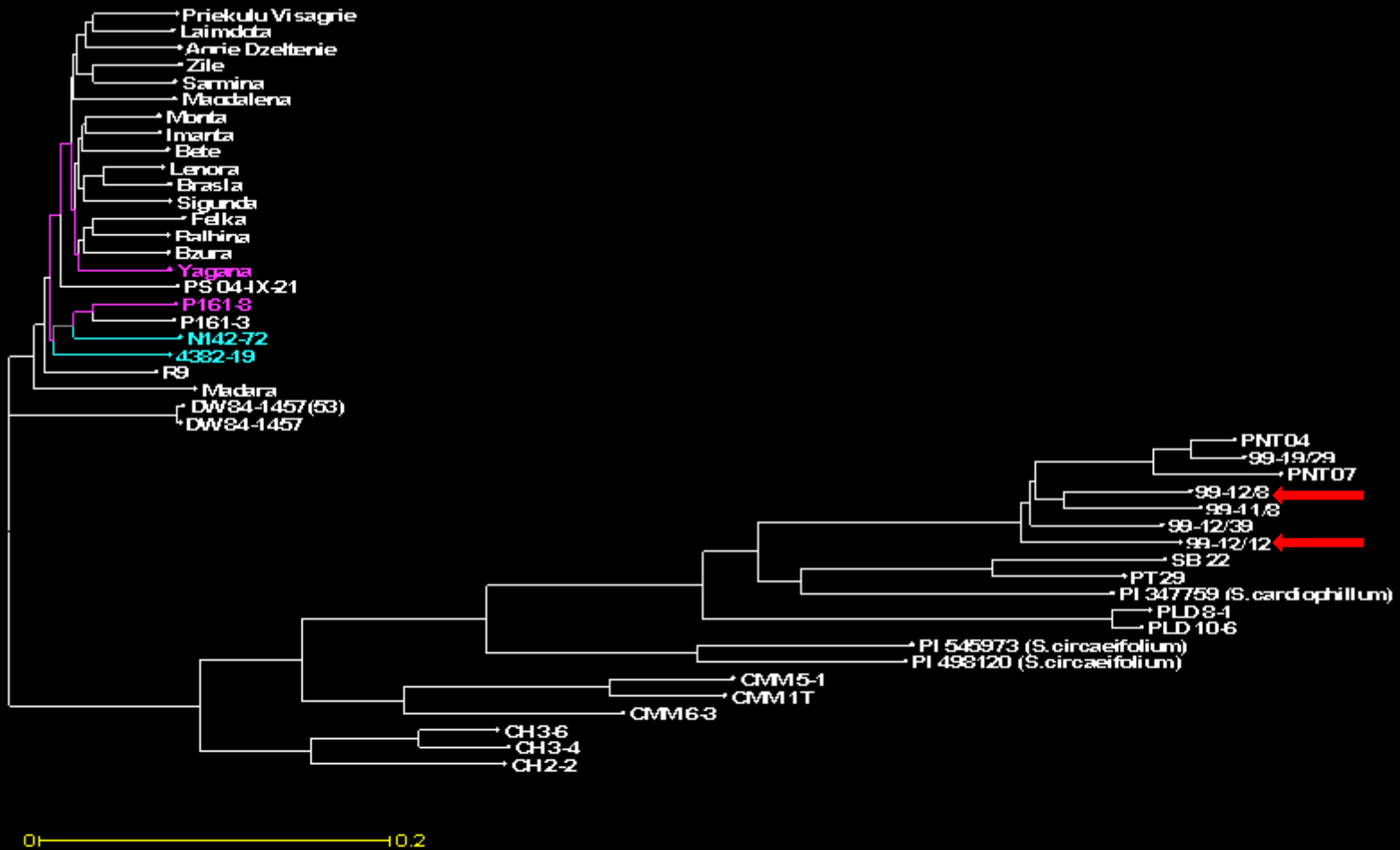


Tuber sizes



Solanum microacanthum

Unweighted neighbour-joining tree based on a Jaccard-distance matrix computed from 1,690 DArT markers



Solanum ruiz-ceballosii Cárđ. 2x (2EBN) **VIR8664 (VIR7370)**

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VII. Series *Tuberosa*

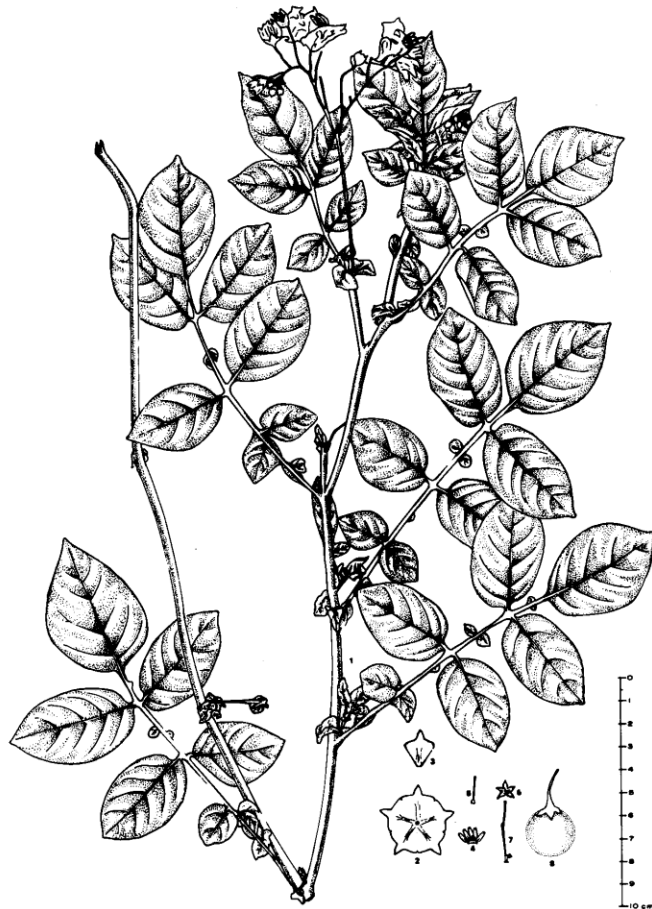


Figure 127. – *Solanum sparsipilum* (Ochoa 662). 1. Flowering plant. 2. Corolla. 3. Petal. 4. Stamens, dorsal view. 5. Pistil. 6. Calyx. 7. Pedicel and pistil. 8. Fruit. All $\times \frac{1}{2}$.

- Originates from:
Bolivia, Peru

habitat: inter-Andean valleys, barren, eroded
or cultivated soil
2400-3000 m above sea level

- Syn. *S. sparsipilum*
- Selected clones:
 - resistant to *P. infestans*
 - low tendency to enzymatic blackening
and after-cooking darkening



Methods for late blight resistance testing

- whole leaf test/spray inoculation *mch*
- detached leaflet test/droplet inoculation *rzc*
- tuber slice test/droplet inoculation *rzc*
- three *P. infestans* isolates
- assessment in 1-9 scale, 9 = most resistant



mch



Mapping populations (R x S)

08-42

- *S. michoacanum* (*mch*)
- 99-12/8 (*mch*) x 99-12/12 (*mch*)
- N = 164
- Resistance tests:
2 dates x 2 rep. X 2 leaves x 6 leaflets
(48 leaflets) in 2009 (MP847)
3 dates x 2 rep. X 1 leaf x 6 leaflets
(36 leaflets) in 2009 (MP921)

05-18

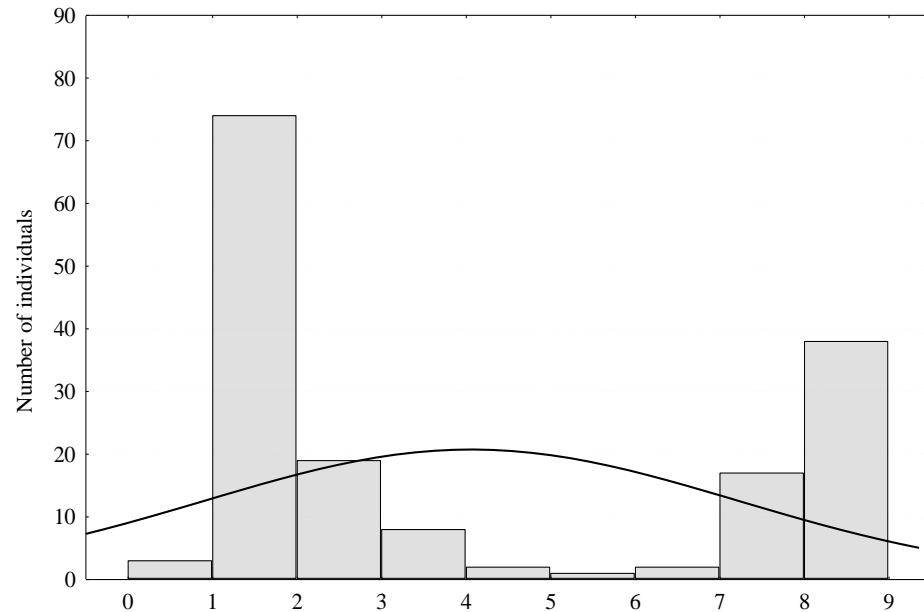
- *S. ruiz-ceballosii* (*rzc*)
- 99-10/36 (*rzc*) x dH Balbina
- N = 114
- Resistance tests (MP 324):
2 dates x 2 rep. x 3 leaflets in 2007
2 rep. x 3 leaflets in 2008
2 dates x 2 rep. x 3 leaflets in 2010

2 rep. x 3 slices in 2006
2 rep. x 3 slices in 2007
2 rep. x 3 slices in 2010

Distribution of mean leaflet resistance in *mch* mapping population

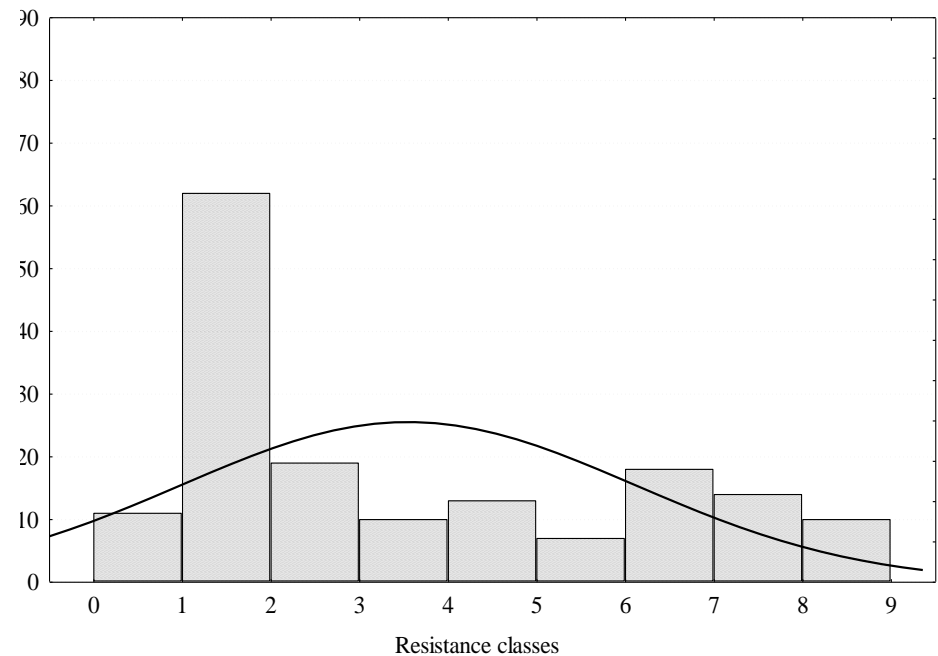
MP 847 (1.3.4.7.10.11) PL 2007

a. 2009
K-S d=0.22, p<0.01



MP 921 (1.3.4.(6).7.10.11) PL 2008

b. 2010
K-S d=0.19, p<0.01





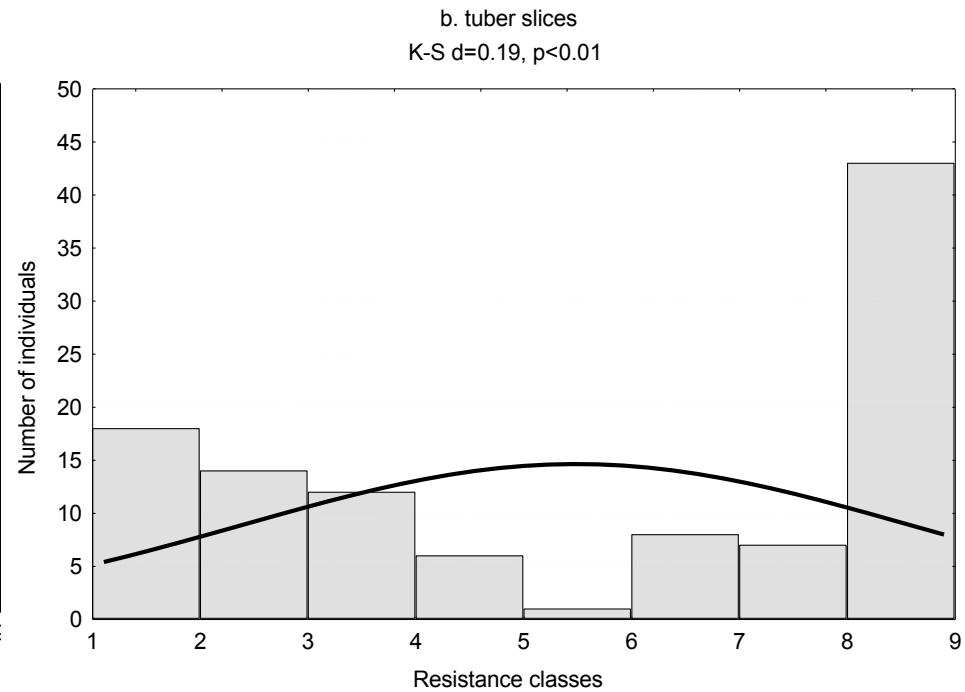
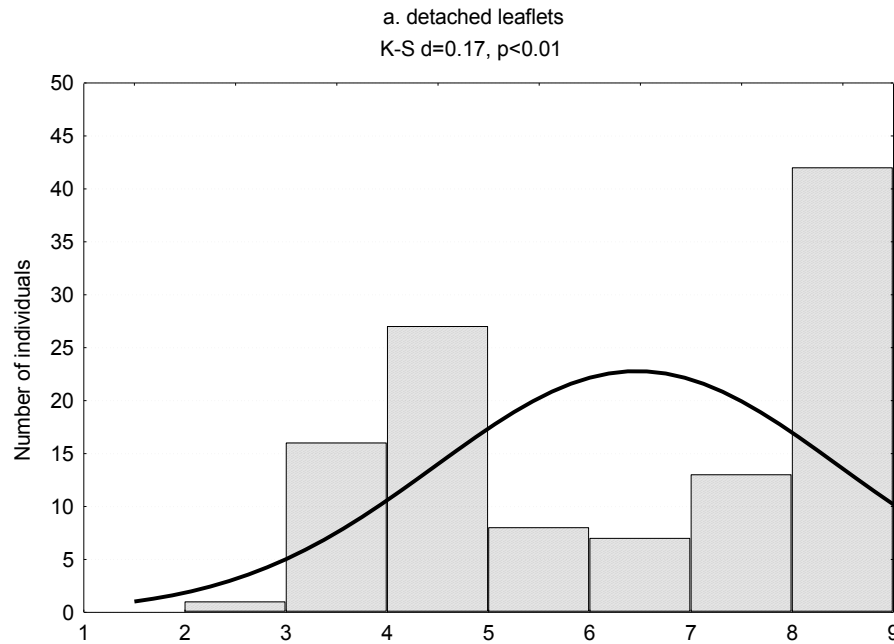
28.05.09
1P
90-103
⑧

mch

Whole leaf test

Distribution of mean leaflet resistance in *rzc* mapping population

MP 324 (1.2.3.4.(5).6.7.(8).10.11) PL 1994



Mapping populations - summary of resistance assessments

08-42

- *S. michoacanum* (*mch*)
- Small leaves and tubers – whole leaf test better than detached leaflet test, tuber tests?
- Pearson's correlation coefficient between results of these two tests: 0.809, $p=0.00$
- R (≥ 6): S ratio 1: 2
- An R gene?

05-18

- *S. ruiz-ceballosii* (*rzc*)
- Pearson's correlation coefficient between results of detached leaflet and tuber slice tests: 0.862, $p=0.000$
- Leaflets R (≥ 7): S ratio 1: 1
- Slices R (≥ 7): S ratio 1: 1
- An R gene
- In leaflet tests - many moderately resistant individuals – additional QTL

Mapping

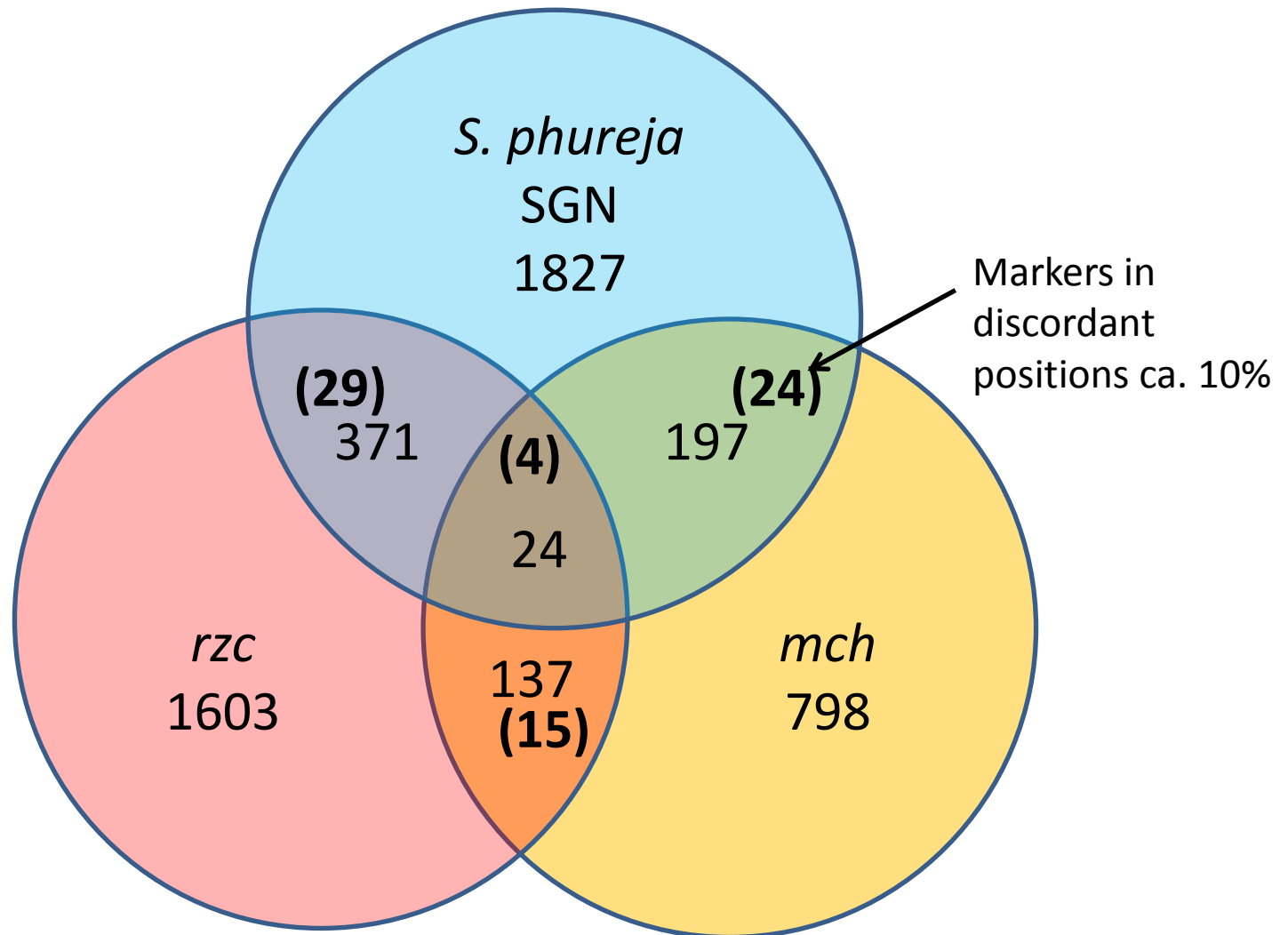
- Diversity Array Technology (DArT)
- Sequence-specific PCR markers for chromosome identification (publication, SGN and GABI databases)
- JoinMap 4[®]: CP (cross pollination) population maternal and paternal maps, common map, independence LOD > 3, regression algorithm and Haldane's mapping function

Mapping Results

Chromosome	<i>rzc</i>			<i>mch</i>		
	Number of markers	Number of reference markers	Length (cM)	Number of markers	Number of reference markers	Length (cM)
I	167	4	103.5	133	4	116.3
II	168	4	113.9	75	4 (+2 ^a)	108.4
III	183	4	143.2	87	4	93.1
IV	74	2	93.9	73	4	99.7
V	195	3	91.8	32	3 (+1 ^b)	66.8
VI	125	3+1 ^a	134.8	74	5	128.2
VII	123	5	82.3	28	4	73.1; 64.1 ^c
VIII	186	5	90.9	99	4	68.0
IX	84	4	107.0	118	4	86.0
X	75	4	79.1	39	4	95.9
XI	124	4	81.8	48	4	54.5
XII	99	5	82.6	40	4	63.0
Total	1603	47+1^a	1204.8	846	48 (+3^{ab})	1047.0

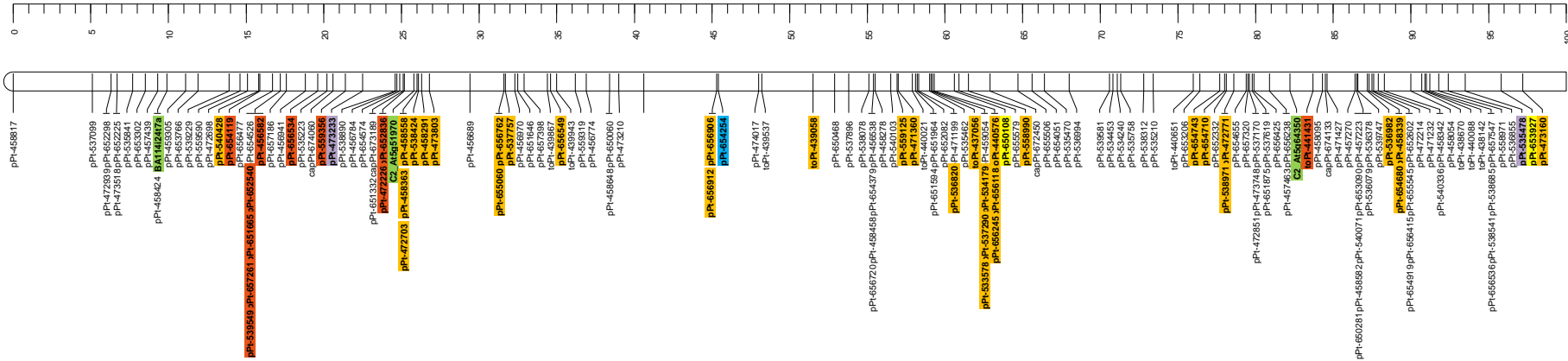
^a markers that on Tomato-EXPEN 2000 and Pepper-COSII maps were located on a different chromosome; ^b markers scored but unmapped; ^cno common map constructed.

DArT maps - comparison

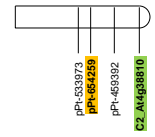


Mapping example: *rzc*

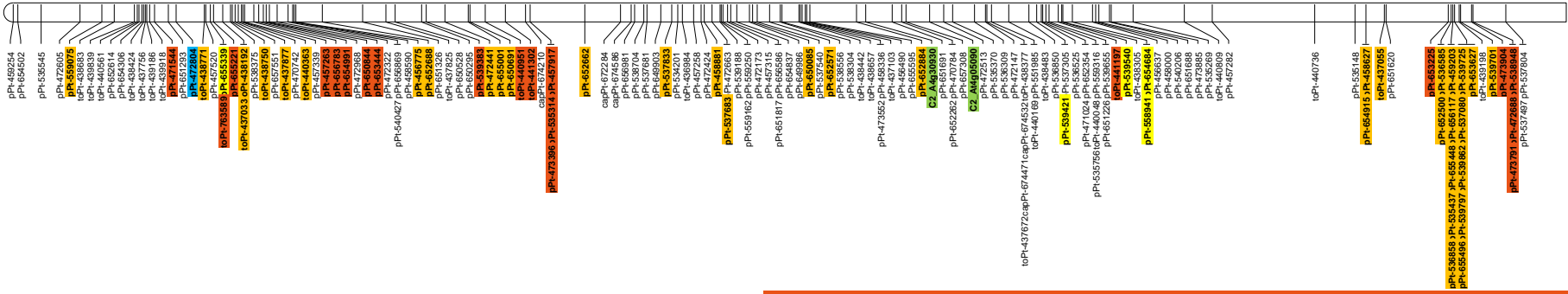
I [1]



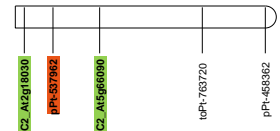
I [2]



II [1]



II [2]



Markers in blue are the markers present on the *Solanum phureja* diploid map 2010 (SGN) that were mapped in discordant positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

Markers in yellow are the markers present on the *Solanum phureja* diploid map 2010 (SGN) that were mapped in corresponding positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

Markers in green are the reference sequence-specific markers.

Markers in orange are the markers present on the *S. michoacanum* map (Śliwka et al. 2011) that were mapped in corresponding positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

Markers in purple are the markers present on the *S. michoacanum* map (Śliwka et al. 2011) that were mapped in discordant positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

Markers in bright yellow are the markers present on both the *S. michoacanum* map (Śliwka et al. 2011) and the *Solanum phureja* diploid map 2010 (SGN) that were mapped in corresponding positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

Markers in red are the markers present on both the *S. michoacanum* map (Śliwka et al. 2011) and the *Solanum phureja* diploid map 2010 (SGN) that were mapped in discordant positions on the dH Balbina x *S. Ruiz-ceballosii* 99-10/36 map.

rzc flower colour

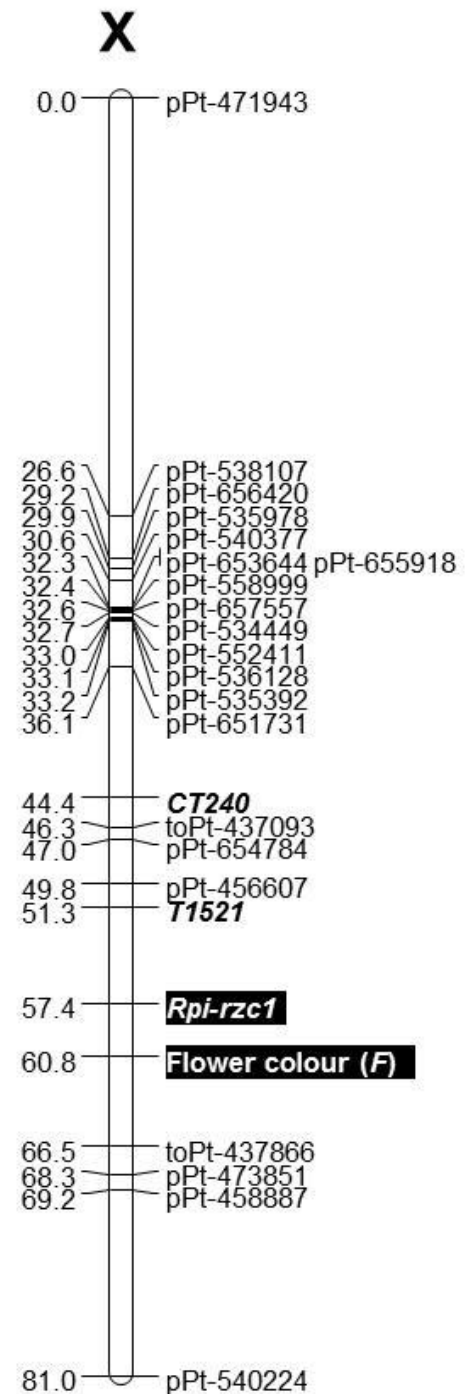
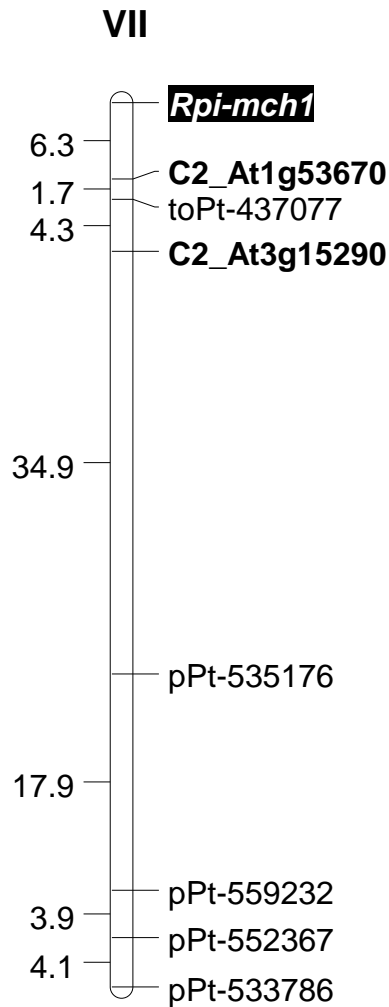
dH Balbina

x

99-10/36 *rzc*



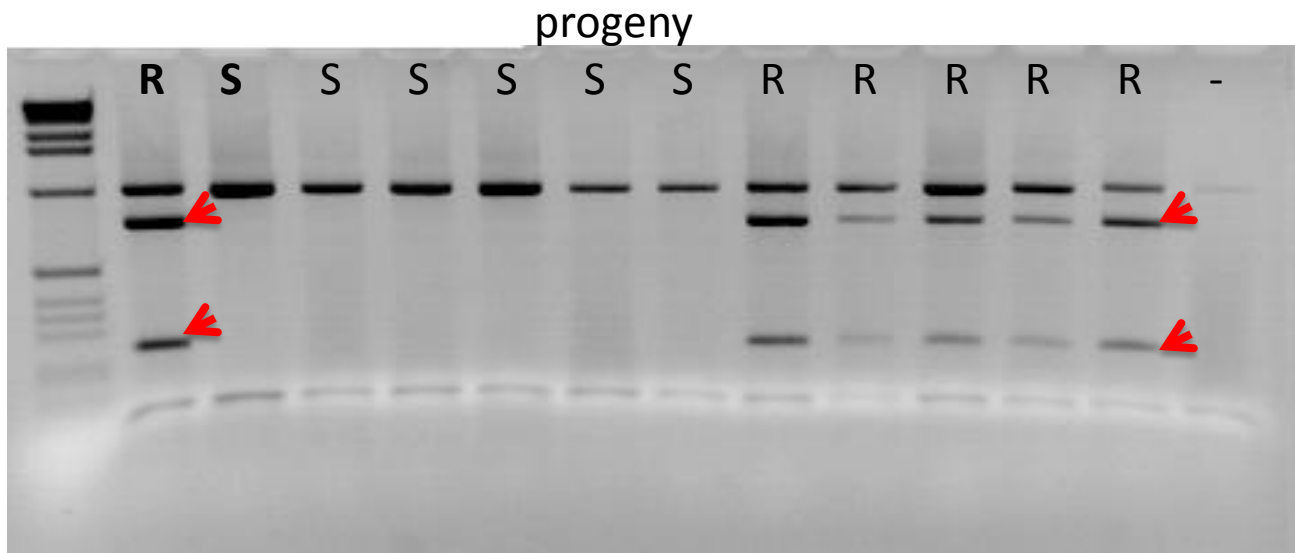
Rpi-mch1 and *Rpi-rzc1*



Rpi-mch1

Marker	P, T student test			R ² (%)		
	Resistance to MP847, 2009	Resistance to MP921, 2010	Weighted mean resistance 2009-2010	Resistance to MP847, 2009	Resistance to MP921, 2010	Weighted mean resistance 2009-2010
C2_At1g53670 <i>RsaI</i>	0.000	0.000	0.000	82.7	67.6	82.2
toPt-437077	0.000	0.000	0.000	75.4	61.4	75.2
C2_At3g15290 <i>TaqI</i>	0.000	0.000	0.000	68.5	54.6	67.4

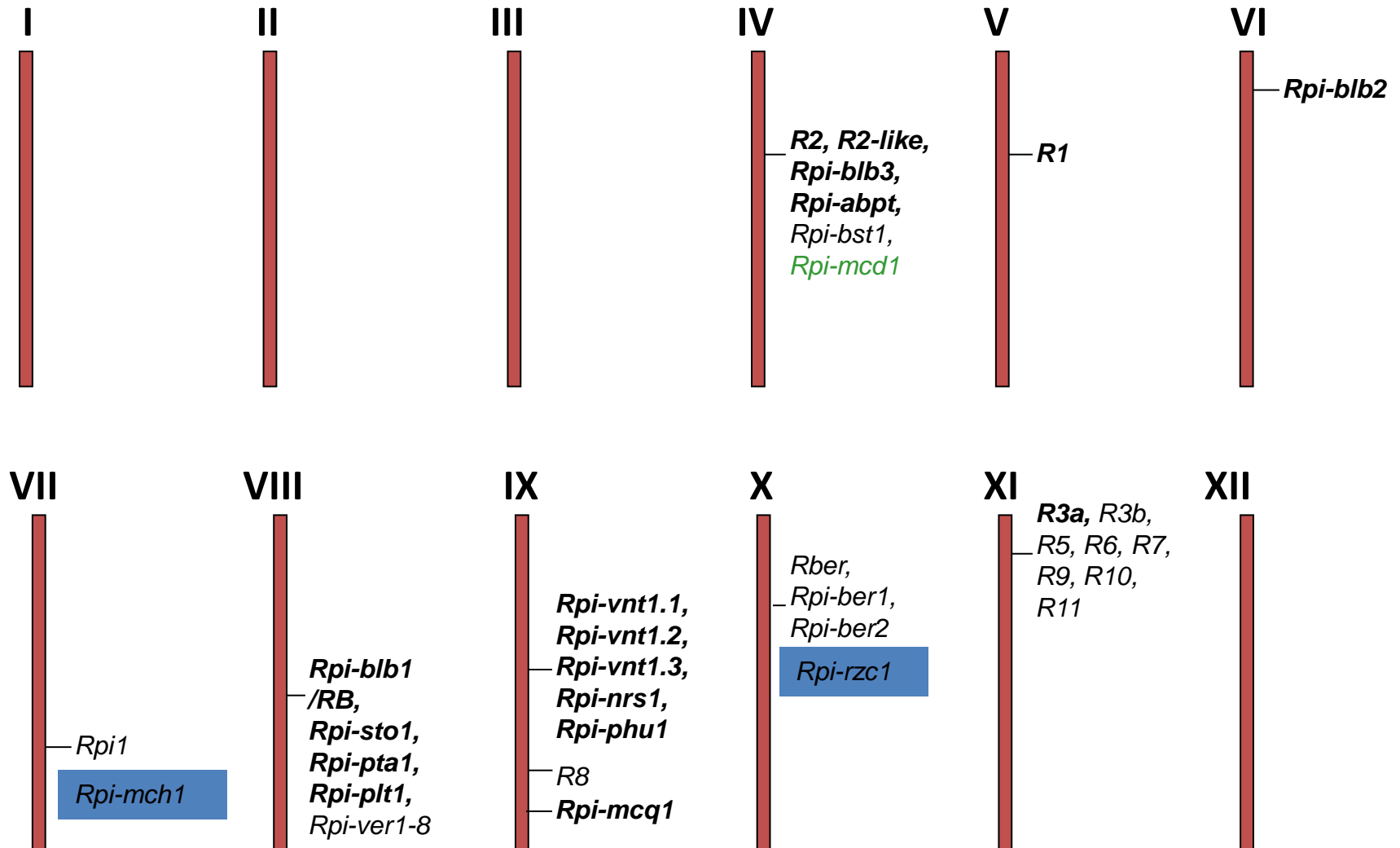
Marker C2_At1g53670, chromosome VII
CAPS, digested *RsaI*



Rpi-rzc1

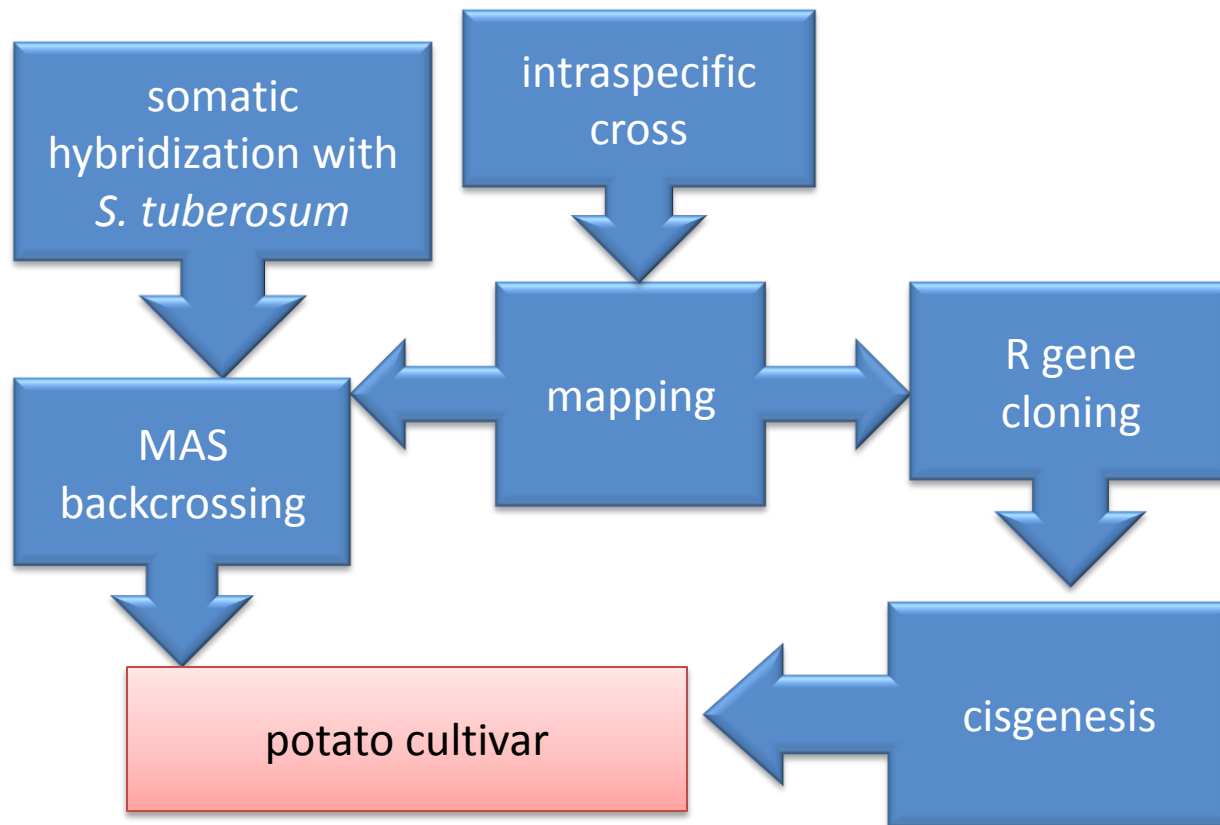
Marker	R ² (%)							
	a. Resistance of detached leaflets				a. Resistance of tuber slices			
	2007	2008	2010	Weighted mean: 2007, 2008 and 2010	2006	2007	2010	Mean: 2006, 2007 and 2010
CT240	58.1	43.0	41.3	54.2	51.0	28.4	36.9	47.2
toPt-437093	53.3	35.3	34.8	46.4	44.8	33.6	32.5	44.8
pPt-654784	69.6	62.3	57.7	73.9	66.9	46.0	54.1	67.8
pPt456607	81.3	49.4	45.0	63.9	63.9	39.6	44.1	59.8
T1521	69.0	43.6	39.8	56.0	52.1	31.1	36.8	48.6
Violet flower colour	70.9	74.7	75.9	87.1	76.9	61.9	75.6	85.7
toPt-437866	70.1	61.8	57.1	73.6	67.3	45.2	53.5	67.4
pPt-473851	52.4	31.7	29.3	41.2	39.7	28.3	27.7	38.4
pPt-458887	50.1	28.4	25.0	36.7	35.7	22.9	23.5	32.9

Hot spots of R genes for resistance to *P. infestans* on potato genetic map



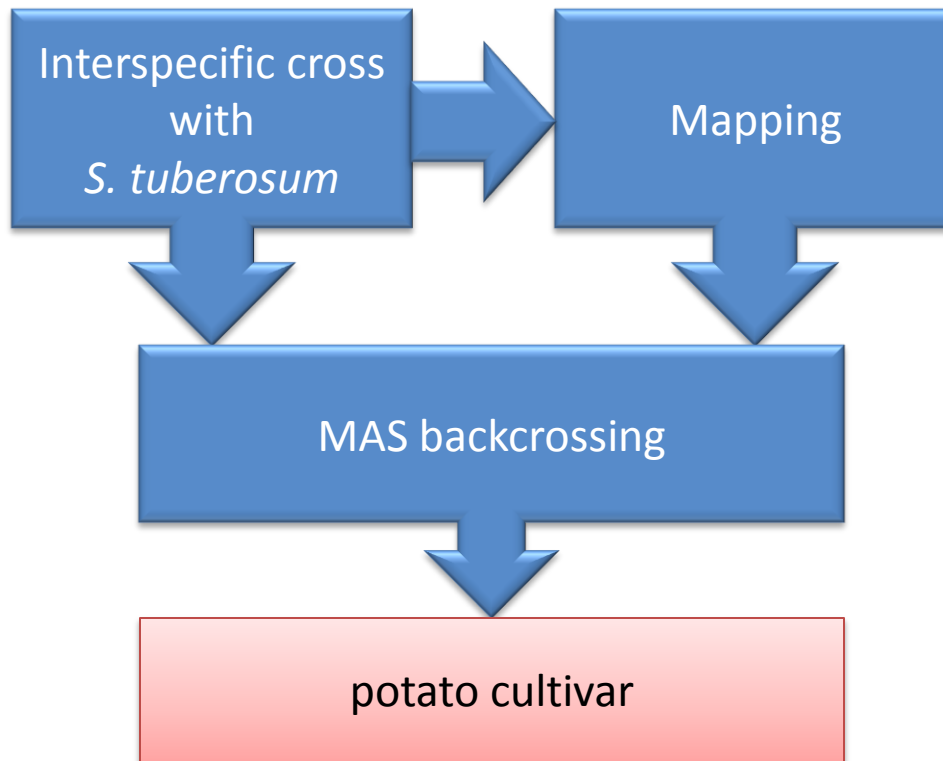
Transfer of resistance to cultivated potato

S. michoacanum

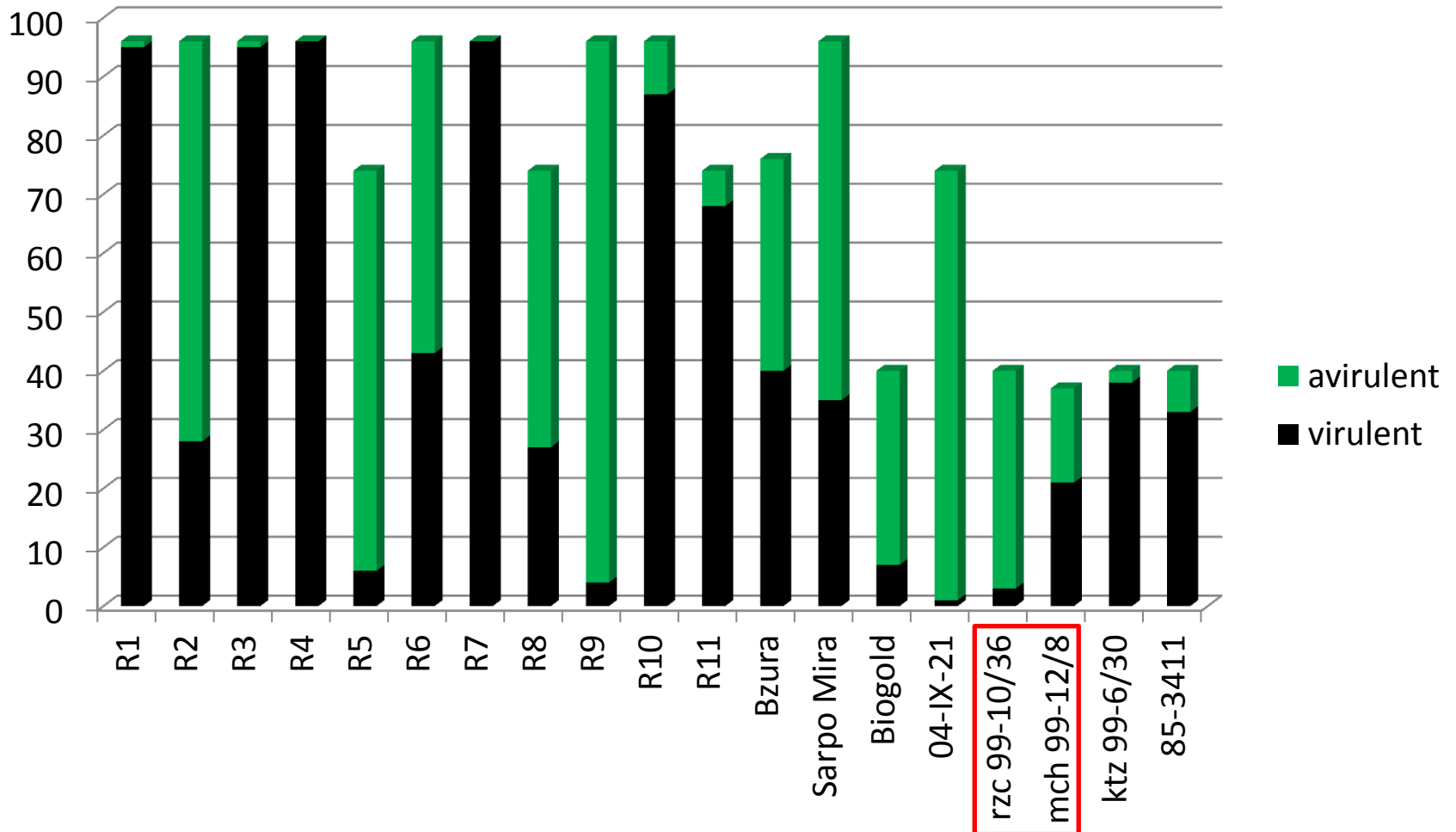


Transfer of resistance to cultivated potato

S. ruiz-ceballosii



Virulence 2009



Summary

- *Rpi-rzc1* and *Rpi-mch1* genes for late blight resistance were identified and mapped
- Both genes can be transferred into cultivated potato gene pool and exploited in gene pyramiding
- Two genetic maps were constructed using DArT markers – valuable resource for new mapping studies



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