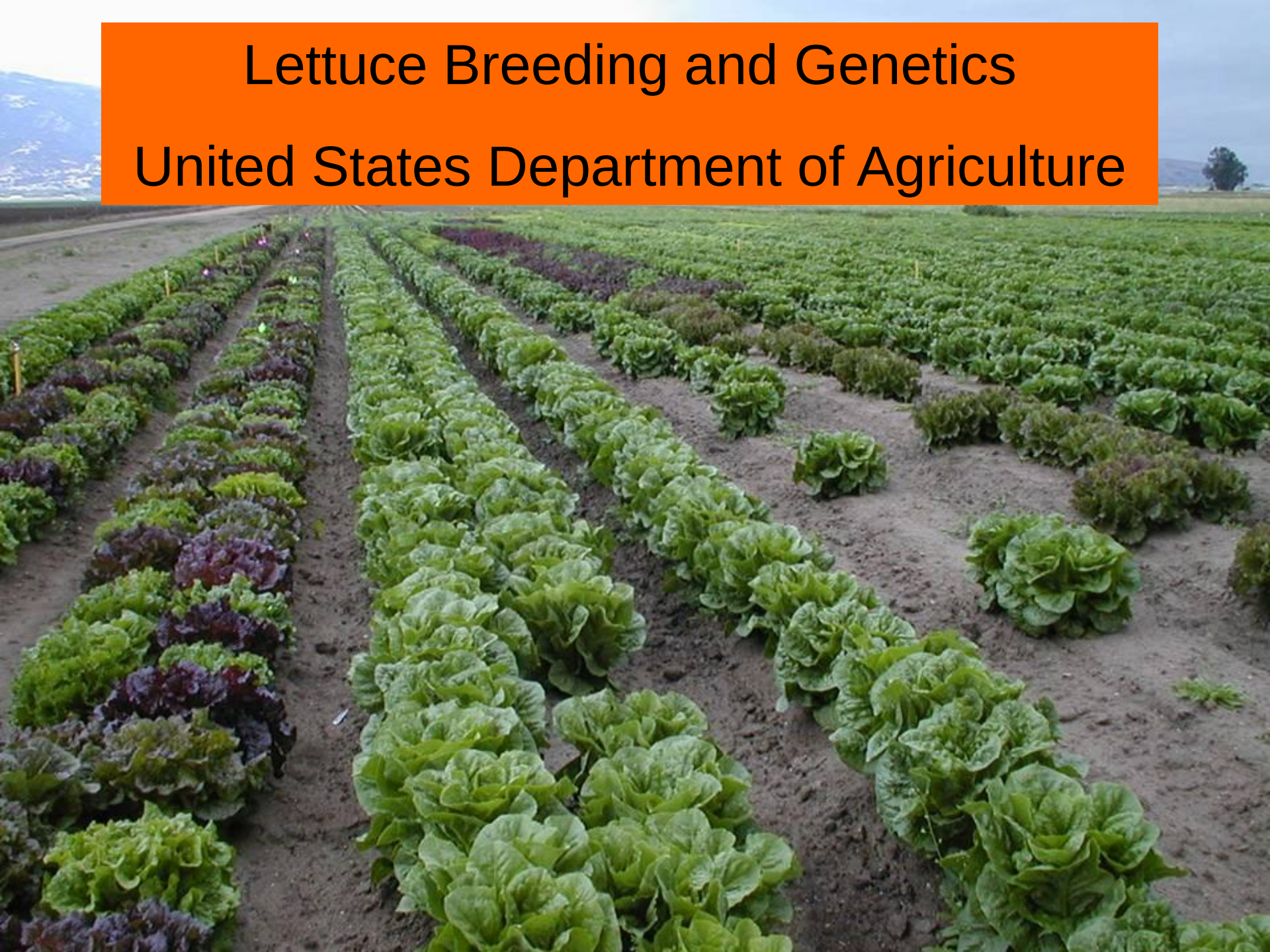
Lettuce Breeding and Genetics

United States Department of Agriculture



U.S. Agricultural Research Station Salinas

Project leaders

- 11 USDA scientist (2 vacancies)
- 3 UC Davis Faculty
- 1 researcher with the Artichoke Research Foundation

U.S. Agricultural Research Station Salinas

- Breeding & Genetics
- Plant Pathology
  - Bacteriology
  - Mycology
  - Virology
- Entomology
- Weed management
- Organic Vegetable Production
- IR-4: Minor Use Pesticides

J.D. McCreight

U.S. Agricultural Research Station Salinas

- Lettuce
- Melon
- Spinach
- Sugarbeet

- Artichoke
- Floral Crops
- Strawberry
- Cover Crops
- Others…

J.D. McCreight

U.S. Agricultural Research Station Salinas

- Powdery mildew-resistant melon
- Monogerm sugarbeet
- ‘Salinas’ lettuce
- ≈40 new viruses
- Sudden Oak Death
- Cover cropping
- Verticillium wilt of lettuce

J.D. McCreight

Lettuce genetic diversity

Origins of lettuce

Lettuce types

- Iceberg
- Batavia
- Romaine
- Latin
- Butterhead
- Leaf
- Stem
- Oil-seed

Lettuce Breeding/Genetics

- *Lactuca sativa* L., *Compositae* family
- 2n=2x=18
- Self-pollinated, cultivars are inbred lines
  - Breeding: pedigree method, SSD, backcrossing
  - Genetics: F_1, F_2, $F_{2:3}$, recombinant inbred lines (RILs)
- 'Salinas' genome sequenced (https://lgr.genomecenter.ucdavis.edu/)
- RIL populations (intra and interspecific)
  - Molecular marker linkage map
  - Numerous trait QTLs mapped (still more to do)
  - Reference linkage map, 13K+ markers. (Truco et al. 2013, G3 4: 617–631)
- Not yet available: MAGIC populations, genome editing

Lettuce Breeding Genepool

L. sativa &
L. serriola (1°)

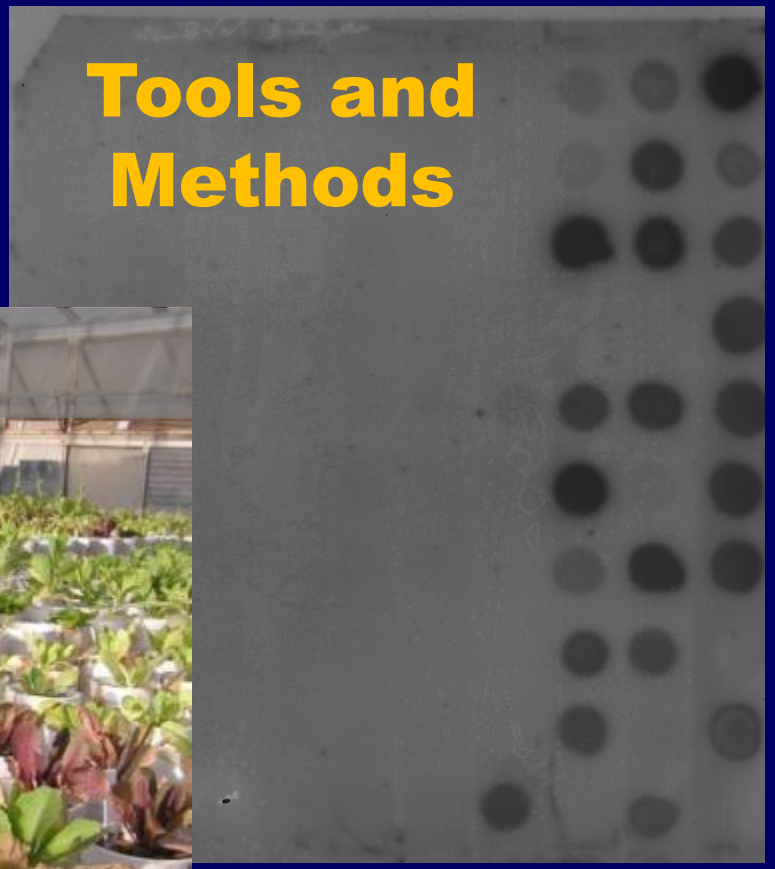
L. saligna (2°)

L. virosa (3°)

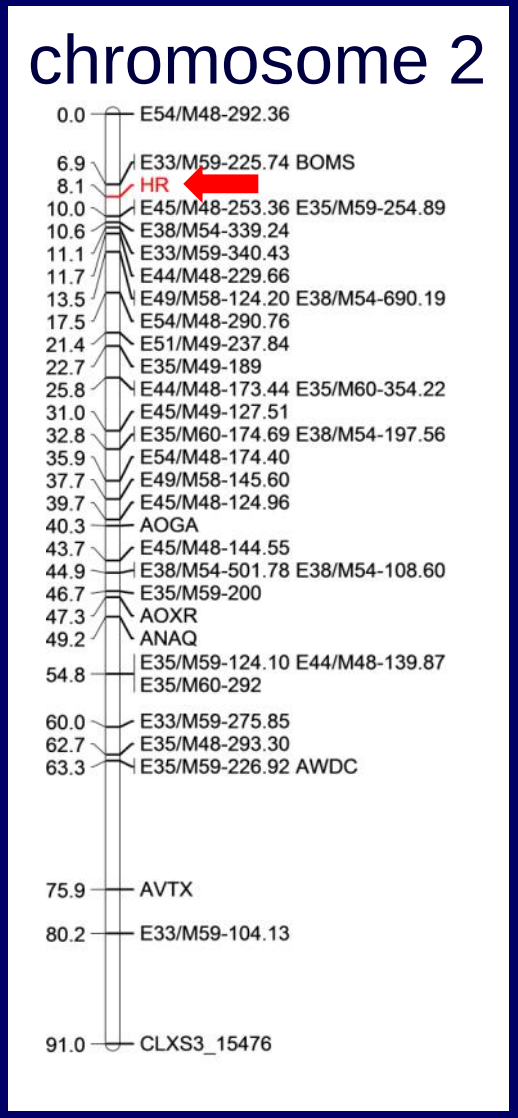
- Genebanks
  - Centre for Genetic Resources, the Netherlands
  - Western Regional Plant Introduction Station, USDA
  - 11,000+ accessions (http://documents.plant.wur.nl/cgn/pgr/ildb/)
    - <10% are wild species

USDA Lettuce Breeding

Lettuce Breeding Goals



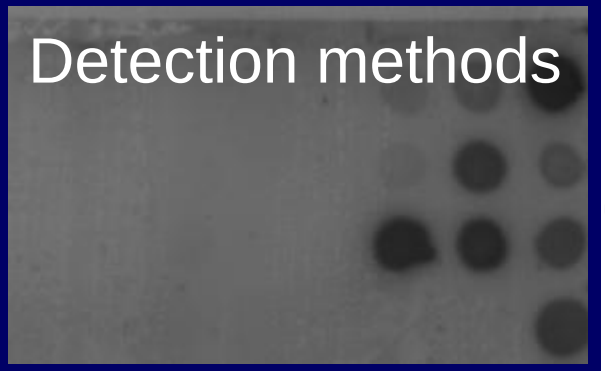
Information: trait genetic diversity and inheritance

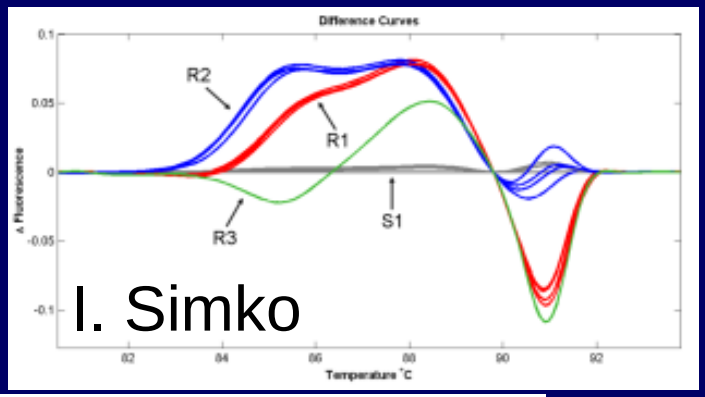


Tools and Methods

Quality Assays

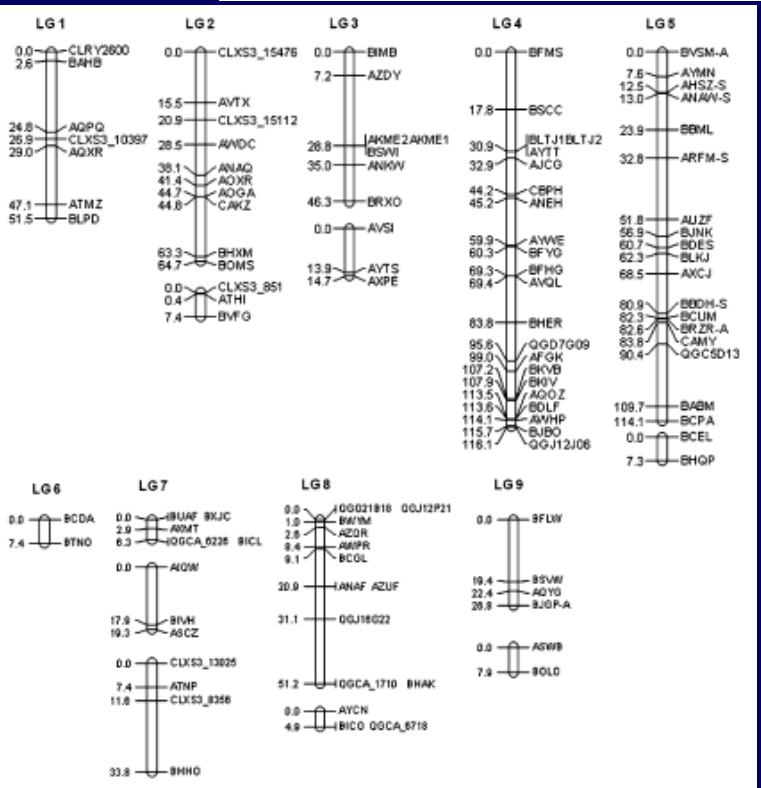
Detection methods





Greenhouse tests

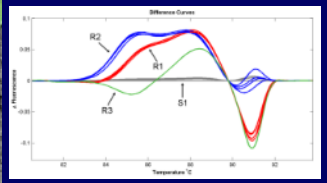
Genetic Resources



GERMPLASM: Characterization of advanced breeding lines

(I. Simko, K. Subbarao, C. Bull, A. Fernandez, Y. Luo)

Resistance: lettuce drop, verticillium wilt, big vein, BLS, downy mildew, dieback, *LMV*



Yield & Quality: tipburn resistance, salad shelf-life, arrival quality

Released Germplasm since 2005 (Mou, Simko, and Hayes)

iceberg & romaine

iceberg

romaine & leaf

leaf

iceberg & leaf

iceberg, romaine & leaf

Lettuce drop resistant romaine, selected from romaine cvr. x Eruption

Eruption

Darkland

-0519

-0488

** sig. less disease than 'Darkland' $P < 0.01$ in two rep. experiments

New breeding schemes

Cross parents of different types


X


- Seed companies select inbreds with uniform type

Evaluate for resistance

- Select early generation families with high and *uniform* resistance
- Variable for plant type

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UNIVERSITY OF CALIFORNIA

Verticillium Wilt of Lettuce

Verticillium dahliae in Lettuce

photos from K. Subbarao

Verticillium wilt management

Broccoli rotation:

- Reduces microsclerotia concentration (Subbarao et al., 2007, Plant Dis. 91:964-972.)

Fumigation and replant to strawberries:

- Provides about 3 crops of protection

(Atallah et al., 2010, Plant Disease, 95: 784-792)

Early harvest of iceberg or grown romaine/leaf

long term goal:

Planting resistant cultivars

Race x Genotype Interaction

Vallad, G.E., et al. 2006. Phytopath. 96:1380-1387.

	V. dahliae isolate	
Lettuce cultivar	Race 1	Race 2
La Brillante Little Gem	Resistant	Susceptible
Salinas Sniper	Susceptible	Susceptible

Race 1 and Race 2 in tomato

- Race 1 and race 2 isolates are also know in Tomato
- An isolate that is race 2 on tomato is also race 2 on lettuce. Same for race 1. (Maruthachalam et al. 2010. Phytopathology 100:1222-1230)

Genetic Variation for Resistance to *V. dahliae* in lettuce

La Brillante

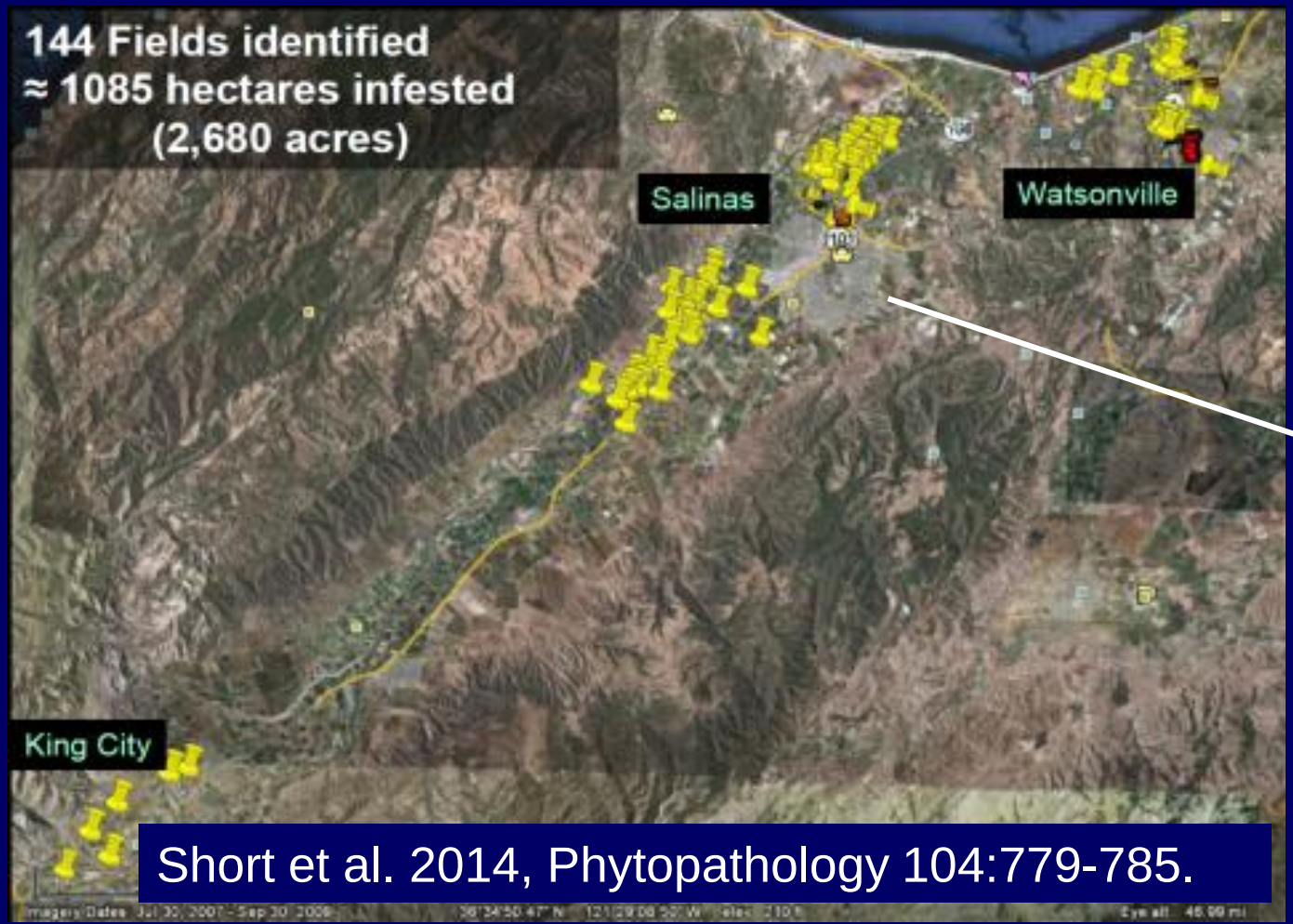
Latin / Romaine

Merlot / Red Leaf

- Race 1 resistance found in wide diversity of lettuce types
- Resistance is complete, no symptoms are observed
- No resistance to race 2
- Race 1 resistance may not be durable

Verticillium wilt distribution

- Salinas Valley isolates tested to date are all race 1
- Fields with race 1 and 2 are known in the Watsonville area



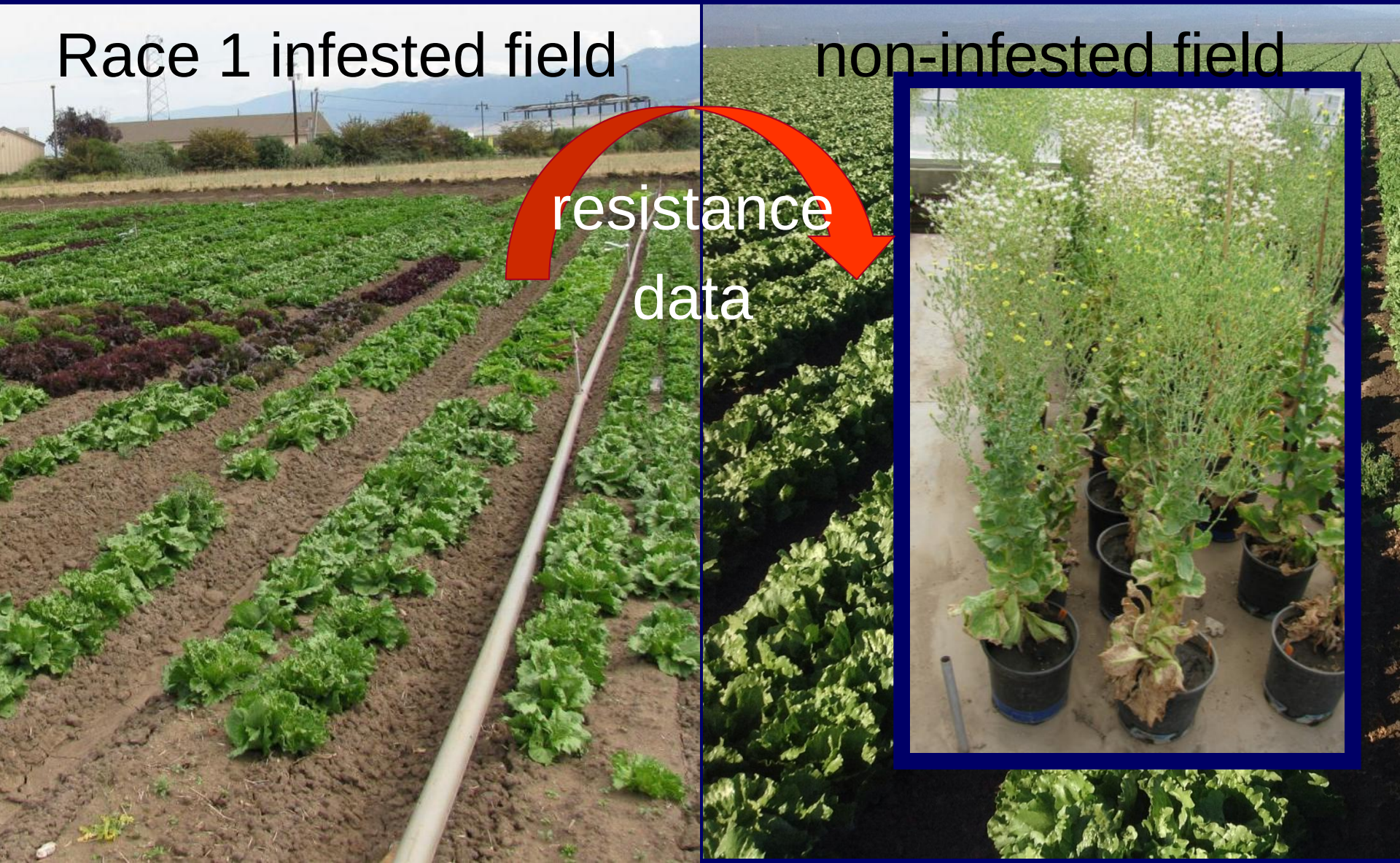
Short et al. 2014, Phytopathology 104:779-785.

GERMPLSM: Field selection for race 1 resistant breeding lines: Pedigree method

Race 1 infested field

resistance data

non-infested field



GERMPLSM: FIELD EVALUATIONS OF THREE F_8 USDA RELEASED BREEDING LINES

Line	Pedigree	2005 Percent Symptomatic	2006 Percent Symptomatic
Pacific		20**	47**
La Brillante		0	5
RH05-0336	Pacific x La Brillante	0	3
RH05-0339	Pacific x La Brillante	0	0
RH05-0340	Pacific x La Brillante	0	0

** Significantly more disease than La Brillante and Breeding lines

Race 1: Iceberg breeding lines with race 1 resistance

Inheritance of Verticillium wilt resistance in other crop species.

- Reports of quantitative resistance in several species (Pegg and Brady, 2002, *Verticillium wilts*)

- Tomato
  - Best described major gene resistance
  - *Ve1*, dominant, race 1 resistance only (Alexander, 1962. Phytopathology 52:998–1000; Kawchuk, et al. 2001 PNAS 98:6511-6515; Fradin, et al. 2009. Plant Phys. 150:320-332.)
  - *Ve1* interacts with *Ave1* (de Jonge, et al. 2012. PNAS 109:5110-5115.)
    - *Ave1* is an effector produced by *V. dahliae*
    - *V. dahliae* without *Ave1* are race 2 and cause disease on *Ve1* carrying cultivars

- Sunflower
  - Single dominant (V_1) and quantitative resistance (Putt, 1958 Crop Science 4:274-276)

Dominance of Resistance to *V. dahliae* Race 1 in F_1 progeny

F_1 Ms7-Salinas x La Brillante

F_1 La Brillante x Salinas 88

Resistance to race 1 *V. dahliae* in F_2 progeny of Salinas 88 x La Brillante

Experiment	No. Plants	Res (NS)	Sus (SYM)	X^2 (p-value) 1 dom. gene (3:1)	X^2 (p-value) 2 dom. genes (9:7)
S88 x LaB F_2 Experiment 1	267	190	77	2.1 (0.15)	59.5 (<0.0001)
S88 x LaB F_2 Experiment 2	97	78	18	2.1 (0.15)	25.0 (<0.0001)
S88 x LaB F_2 Total	363	268	95	0.2 (0.70)	52.7 (<0.0001)

NS = Non symptomatic, Res = Resistant

SYM = Symptomatic, Sus = Susceptible

INFORMATION:

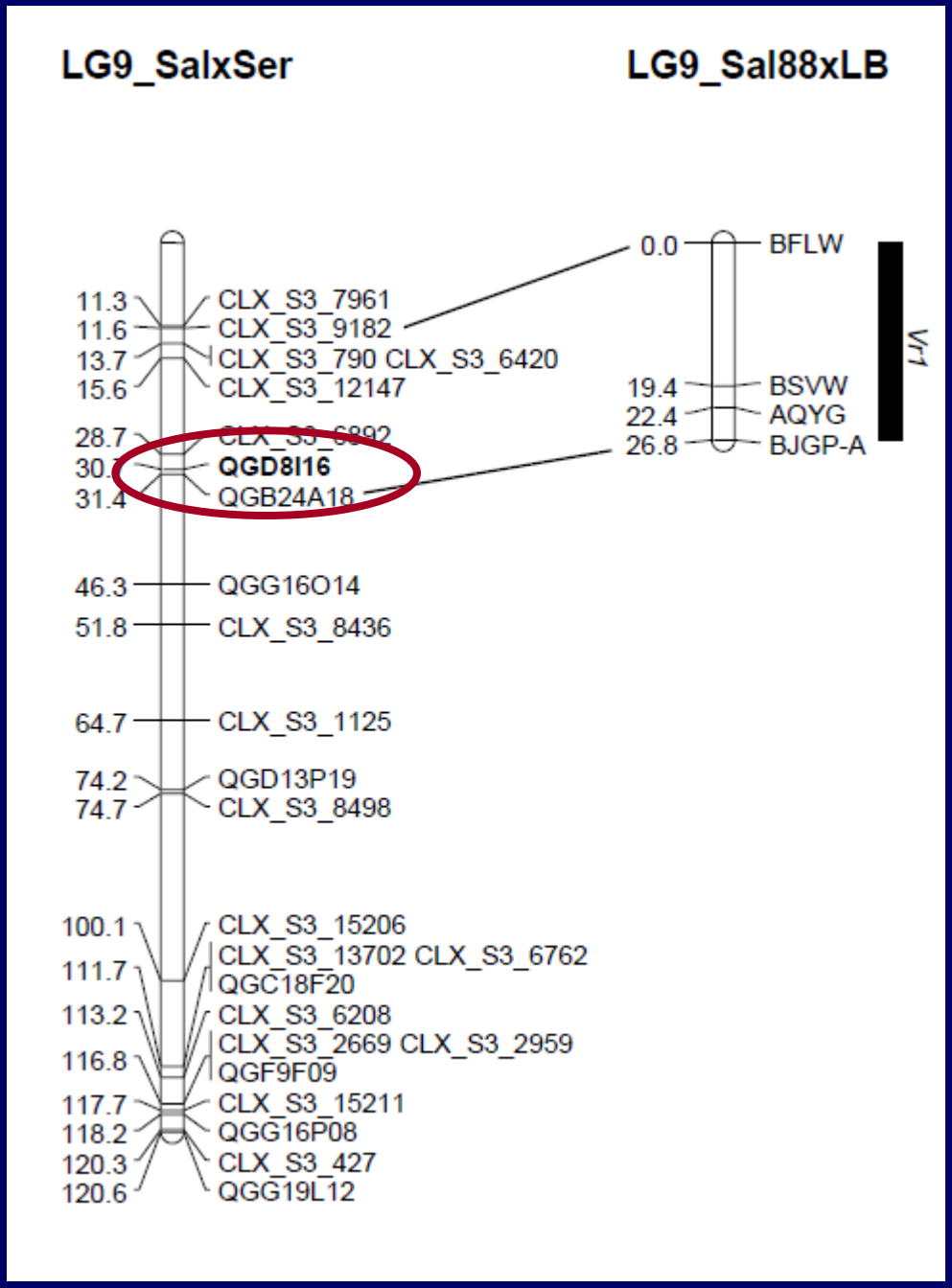
Inheritance of Verticillium wilt resistance in lettuce

(w/ Maria Truco and Richard Michelmore, UCD)

Resistance locus, named *Vr1*, positioned on linkage group 9

Coincident with marker QGD8I16, which has sequence similarity to the *Ve1* in tomato

This is a good candidate to be the gene conferring resistance to race 1



Inheritance of resistance to race 1 Verticillium wilt in lettuce: what does it all mean

- Race 1 resistance is simply inherited
  - Backcrossing could be an effective breeding scheme for this trait
  - *Potentially* faster and easier than pedigree breeding
- Resistance in Tomato and Lettuce may be functionally similar.
- Lettuce genes with *Ve* like sequences are good targets for functional testing and development of molecular markers
  - Marker assisted selection could accelerate backcrossing breeding

Race 2

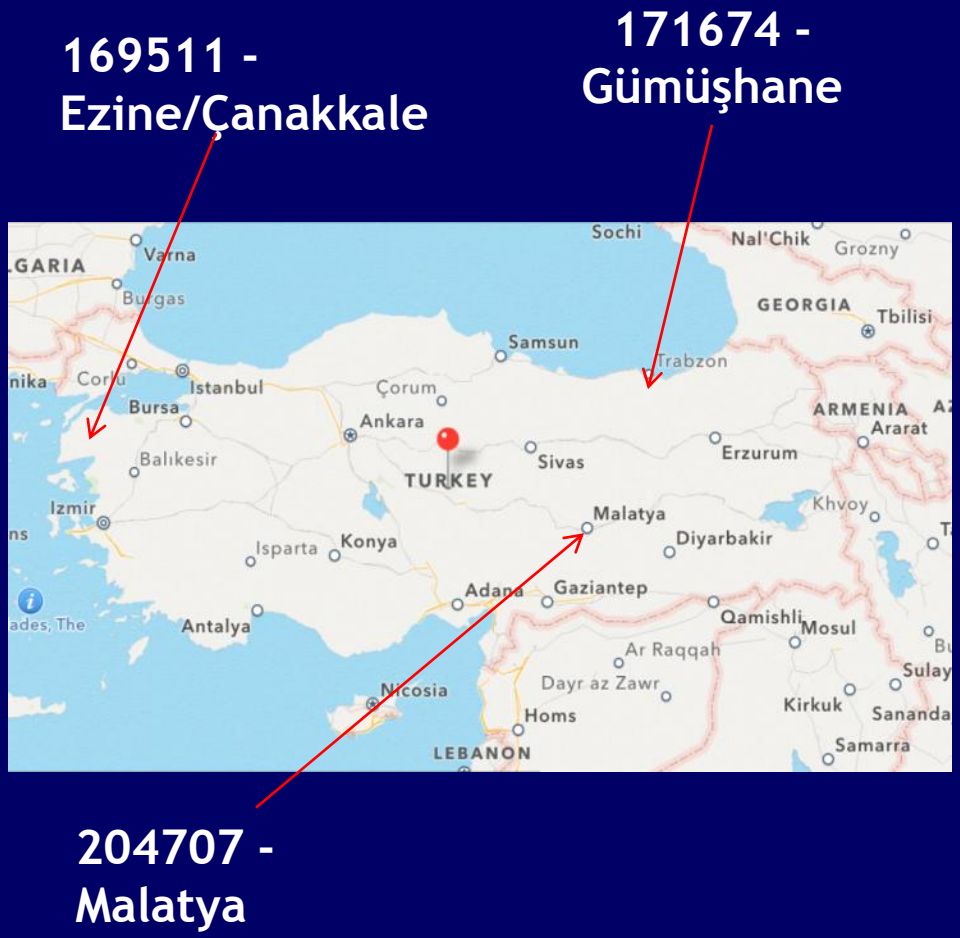
- Race 2 Isolates cause disease on La Brillante and all other cultivars with complete resistance to race 1.

- Race 2 resistance research primarily conducted by German Sandoya

No field site infested with race 2

New sources of resistance to race 2 of *V. dahliae*

- Evaluating USDA, Western Regional Plant Introduction Station collection
- Only partial resistance found: PIs 169511, 204707, 226641 and 171674 (Hayes et al, 2011)
- All *L. sativa* landraces
- No complete resistance known
- Screened 850+ accessions so far, is an ongoing project



Germplasm collecting through the USDA plant exchange office (K. Tamanyan, M. Mosulishvili, S. Litvinskaja, R. Murtazaliev A. Asgarov, N. Quliyev, M. Eldarov)

Completed explorations

Armenia: 2009

Georgia: 2009 & 2010

Russia: 2010

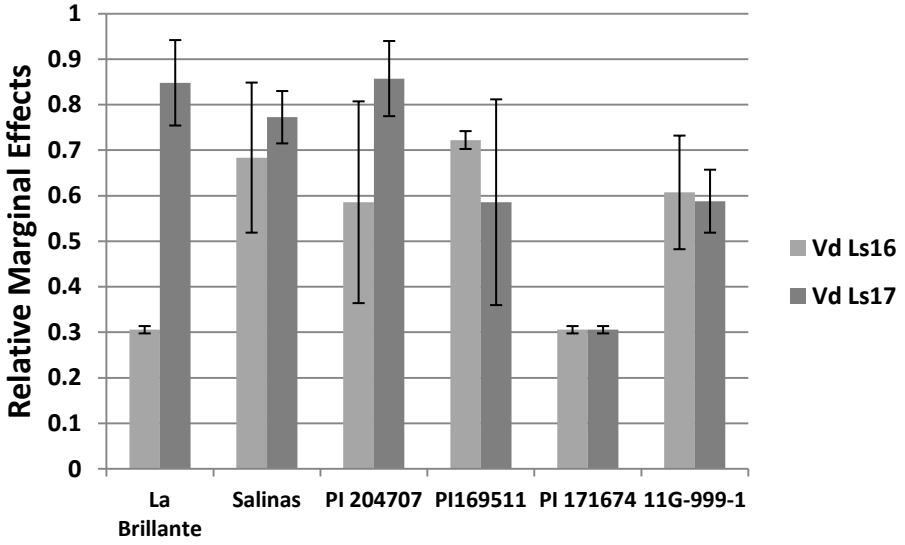
Azerbaijan: 2014

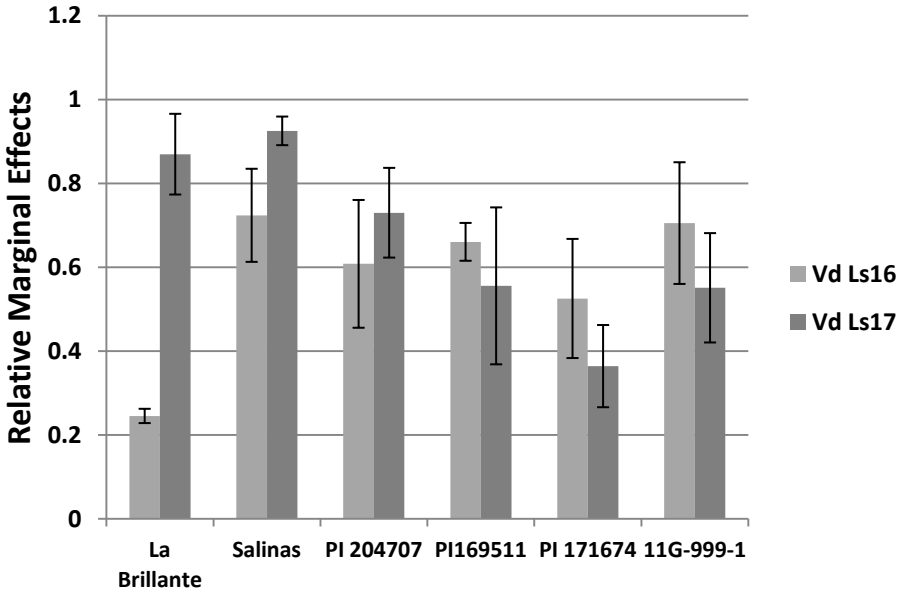
Possible Collecting

Turkey

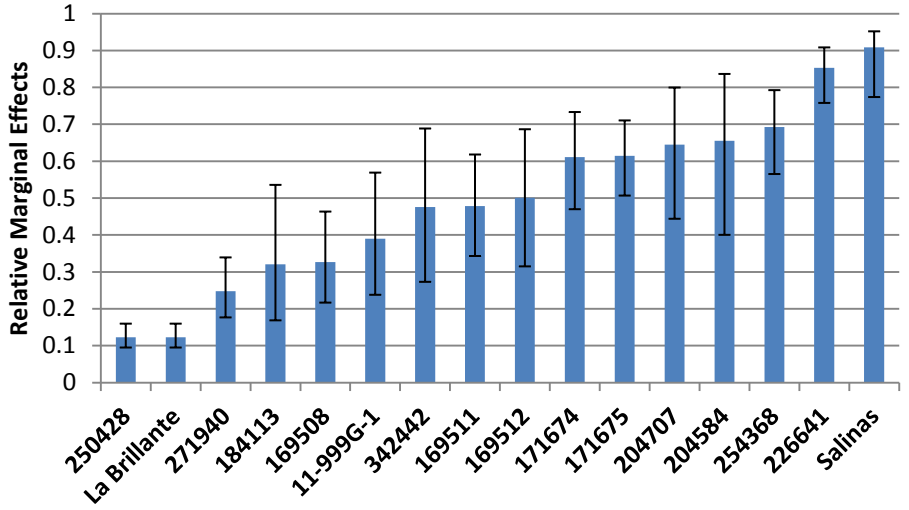
Resistance to Race 1 and Race 2 (G. Sandoya)

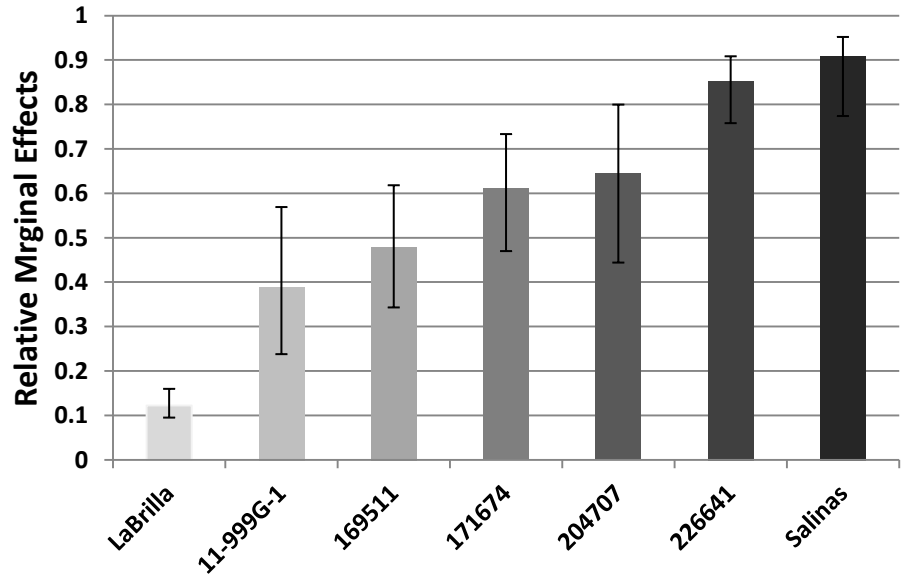
Race 1 & 2 Greenhouse experiment





Race 1 ONLY! Field experiments 2012 and 2013

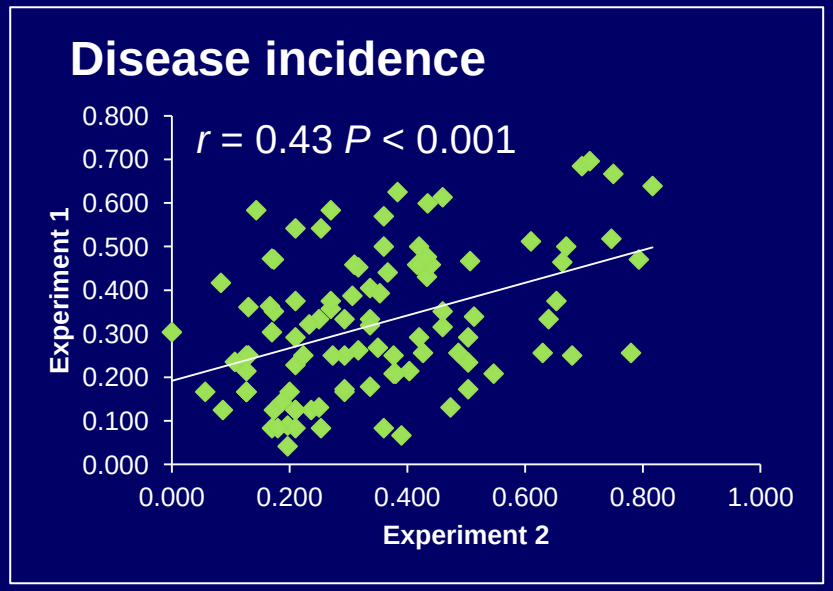




Race 2: Populations for race 2 breeding and genetics (G. Sandoya)

Cross	Phenotype	Generation	No. of families
PI 169511 x Salinas	**PR. x S.**	**F3**	**169**
L. ser 11G999-1 x PI251246 (UC popl'n)	**S x PR**	**F7**	**256**
PI 171674 x PI 204707	**PR. x PR.**	**F3**	**198**
Salinas x PI 171674	**S. x PR**	**F3**	**174**
PI 171674 x L. ser 11G999-1	PR. x PR.	F3	250
(F_1 - Salinas x PI 171674) x RH12-3196	Race 2 x Race 1 res	F2	260
PI 169511 x PI 204707	PR. X PR.	F3	194
PI 226513 x PI 206965	PR. x PR.	F3	187
PI 179295 x PI 169511	PR. x PR.	F3	198
Salinas x PI 226513	S. x PR.	F3	183
Salinas x PI 206965	S. x PR.	F3	167
PI 169511 x PI 171674	PR. x PR.	F3	199
PI 274366 x PI 169511	PR. x PR.	F3	198
PI 171674 x PI 226641	PR. x PR.	F2	na
PI 169511 x L. ser 11G999-1	PR. x PR.	F2	na
PI 169511 x PI 206965	PR. x PR.	F2	na
PI 204707 x L. ser 11G999-1	PR. x PR.	F2	na
PI 226641 x L. ser 11G999-1	PR. x PR.	F2	na
PI 169511 x PI 204707	PR. x PR.	F2	na
Salinas x L. ser 11G999-1	PR. x PR.	F2	na

Race 2: Segregation of F_3 families from PI 171674 x PI 204707 in two replicated greenhouse experiments

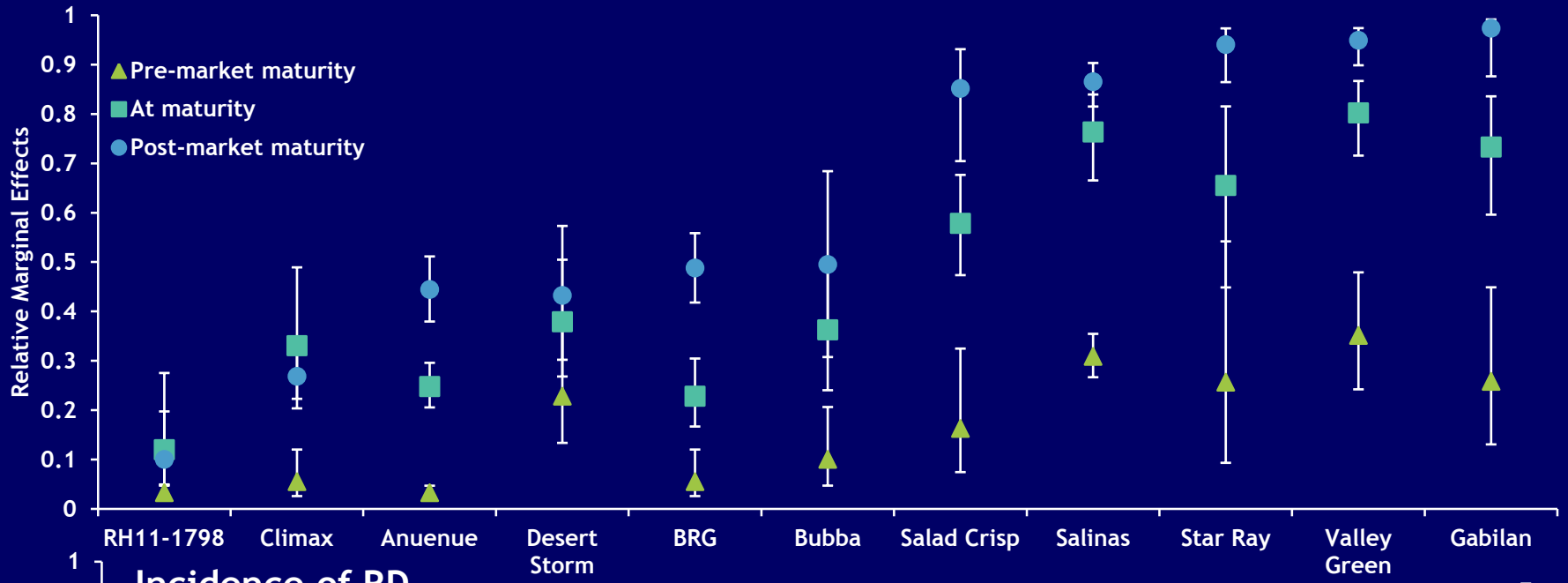


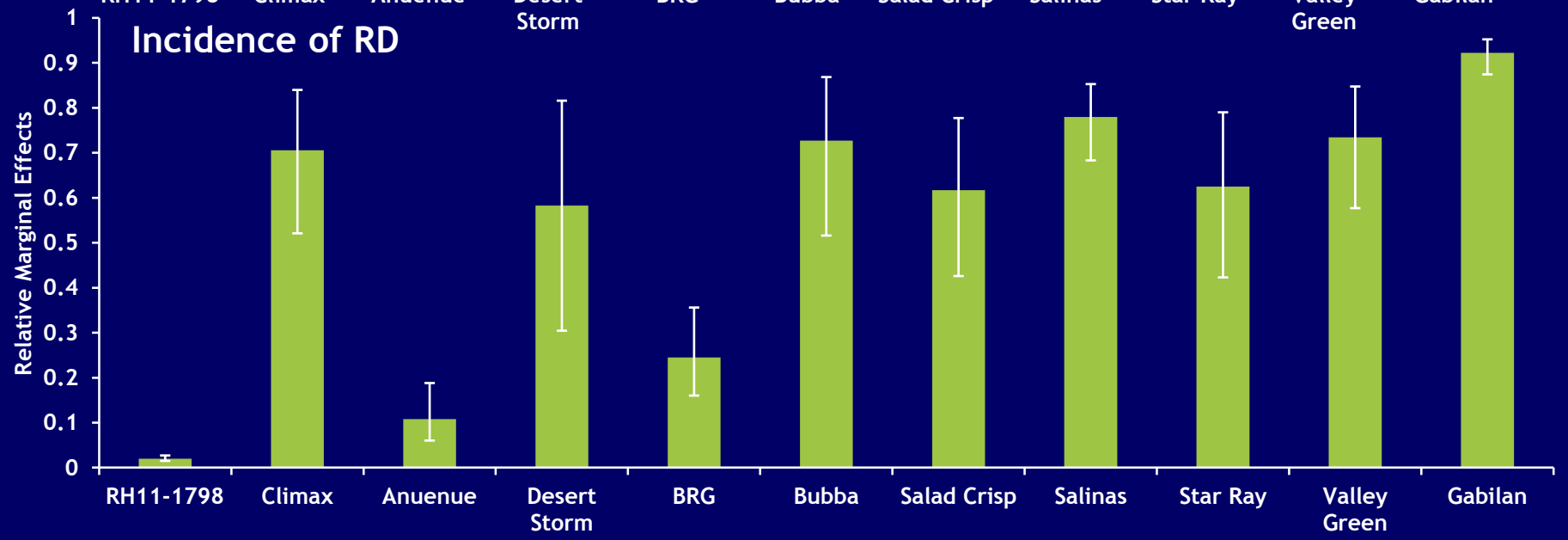
- Resistance is quantitatively distributed
- Correlation between experiments was significant but of moderate magnitude
  - Indicates segregation for resistance
  - Environment plays a large role in disease
  - Race 2 resistance breeding probably more challenging than race 1

Delayed foliar symptoms (G. Sandoya)

- 34 iceberg lettuce cultivars adapted to different conditions
- Race 1-infested field
- Three expts with three replicates
- Evaluate disease & maturity at three time-points
  - Pre-market maturity
  - At market maturity
  - Post-market maturity

Delayed foliar symptoms: results (G. Sandoya)





VERTICILLIUM WILT RESISTANCE BREEDING & GENETICS: FUTURE DIRECTIONS

- Race 1
  - Continued breeding of race 1 resistant icebergs
    - Combine *Vr1* with resistance to downy mildew and cork root
  - Marker development
- Race 2
  - Finish genetic analysis of partial resistance/delayed wilting
    - Is the level of resistance sufficient?
    - Combine genes from multiple sources of resistance
    - Is breeding with partial resistance/delayed wilting feasible in practice?

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