WG1: Vegetable rootstock breeding: genetic variation and selection strategies

Prof Andrew Thompson

19th September, Pula, Croatia

www.cranfield.ac.uk

Working group 1 – summary of activities

1. ~7 international meetings – breeders, experts in germplasm and molecular genetics => collaborations

2. Contribution to Veg. Grafting training school Catania: genetic resources and breeding (AT, Enza Zaden, Rijk Zwaan)

3. Visits to breeding companies with interests in vegetable rootstocks

4. Book chapters:

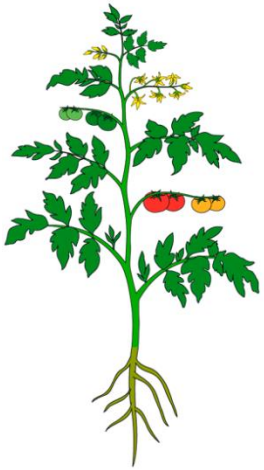
Chapter 2: Genetic resources for rootstock breeding
Belen Pico, Andrew Thompson, **Carmina Gisbert**, Halit Yetisir, Penelope Bebeli

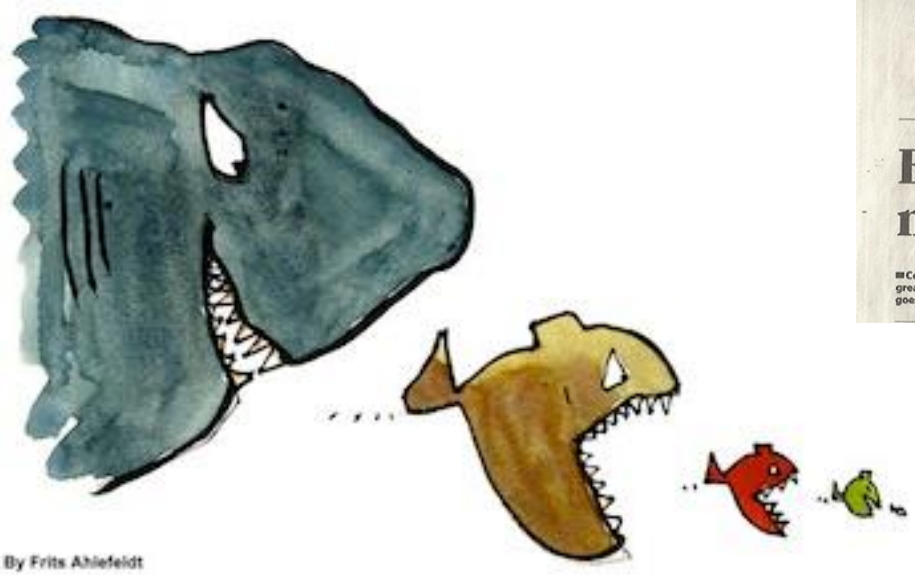
Chapter 3: Rootstock breeding: current practices and future technologies
Andrew Thompson, Belen Pico, Halit Yetisir, **Roni Cohen**, Penelope Bebeli

WG1: Engagement with seed companies

Company/Group[1]	Attended meetings	Visits (group or individual)
Asahi Industries		
Bejo Zaden		
BHN Seed		
Capgen Seeds		
De Ruiter/Monsanto	✓	
DP Seeds		
Enza Zaden	✓	✓
Gautier Semences		
Hazera/Limagrain		✓
Vilmorin/Limagrain	✓	
Nunhams/Bayer	✓	
Origene		
Rijk Zwaan	✓	✓
Ramiro Arnedo Semillas	✓	✓
Sakata		
Seminis/Monsanto	✓	
Syngenta	✓	✓
Takii Seed		
Unigenia	✓	✓
Zeraim Gedera/Syngenta	✓	✓

Market-leading tomato rootstocks Maxifort and Beaufort are coming home to EU, now German owned!





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THE SACRAMENTO BEE

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Bayer-Monsanto deal may cut ag jobs here

■ Companies have agricultural technology operations in Davis and greater Sacramento. If Bayer's $66 billion purchase of Monsanto goes through, duplicated operations might be eliminated.

ations in Davis have their roots in the earliest rumblings of biotech in the region, going back to a genetically modified

santo's shareholders.

The impact on the two companies' operations in Davis, Woodland and other Northern California communities remains to be seen. A Bayer spokesman at the company's headquarters in Germany said it's too early to predict.

At the same time, though, Bayer said it expects to squeeze $1.5 billion in "total synergies" from the merger over three

CAPITOL & CALIFORNIA

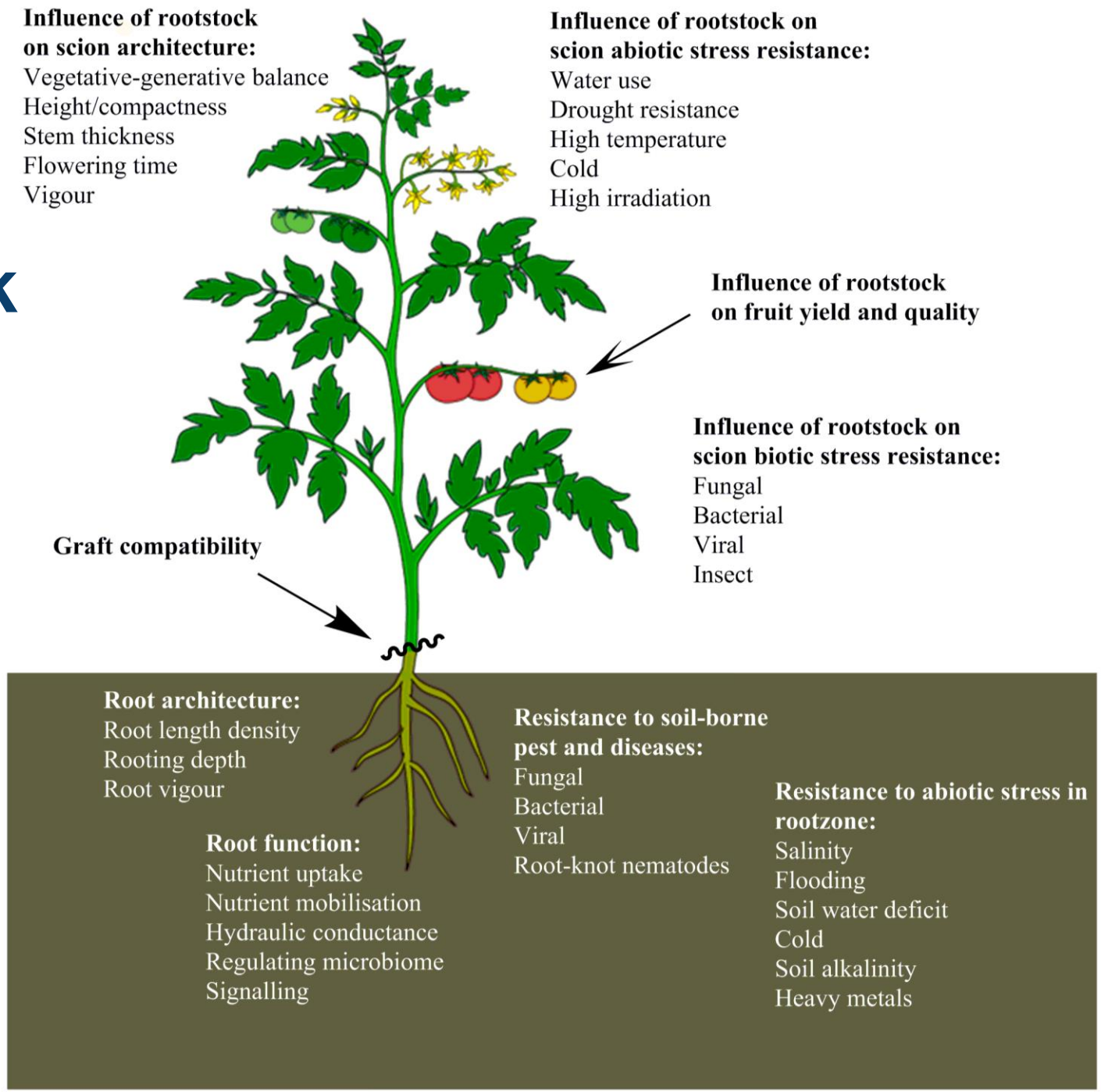
BROWN BEGINS CAP-TRADE PUSH

The governor signed a bill authorizing $900 million for climate-related programs, including parks and public transit. 7A

Rootstock traits

WP2
WP3
WP4

Book Chapt 3

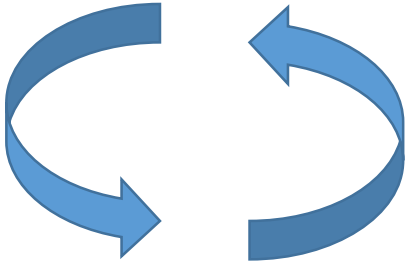


Plant Breeding – What is it?

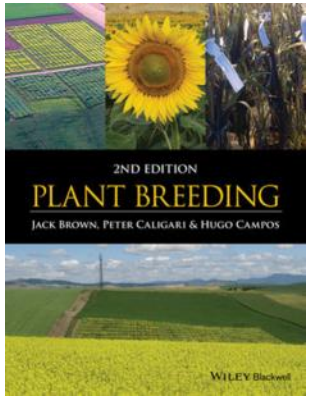
"…to develop superior cultivars…..adapted to specific environmental conditions and suitable for economic production in a commercial cropping system"

1. produce or identify genetically variable germplasm
2. select superior genotypes with specified characteristics (growers, processors and consumers must be happy)
3. stabilize and multiply for commercial release

1. Increase diversity



2. Decrease diversity



The impact of grafting on breeding strategies

Trait stacking is challenging:

1. Mathematics of independent assortment and problems of “linkage drag”

2. Epistatic interactions. The action of one gene may depend on a specific allele at another locus.

3. Pleiotropic effects: genes that affect rootstock may have negative effects on the shoot, if expressed in non-grafted plants.

4. Complex quantitative traits – large number of loci may have a small influence on a trait

The impact of grafting on breeding strategies

Trait stacking is challenging: BUT:

- grafting breaks down one big problem into two easier-to-solve problems

Which squirrel would you rather be?

The impact of grafting on breeding strategies

Grafting makes it easier to deliver improved root traits

1. Adding root traits to elite germplasm by crossing requires reselection of all shoot traits (~10 years). Faster to break one big problem down into two smaller problems with different objectives

2. Roots traits interact strongly with soil conditions that can vary between fields. Quicker to deploy different rootstock × scion combinations than to locally adapt non-grafted cultivars. **But not done in practice!**

3. Rootstocks allow deployment of a lot of wild species DNA with far less chance of reducing fruit yield and quality.

4. Potential pleiotropic effects can be avoided.

Mendelian genetics: divide and conquer

Grafted, 2 + 2 genes

1 in 16 scion

Cross:
AaBb × AaBb

	AB	Ab	aB	ab
AB	AABB	AAbB	aABB	aAbB
Ab	AABb	AAbb	aABb	aAbb
aB	AaBB	AabB	aaBB	aabB
ab	AaBb	Aabb	aaBb	aabb

1 in 16 rootstock

Cross:
AaBb × AaBb

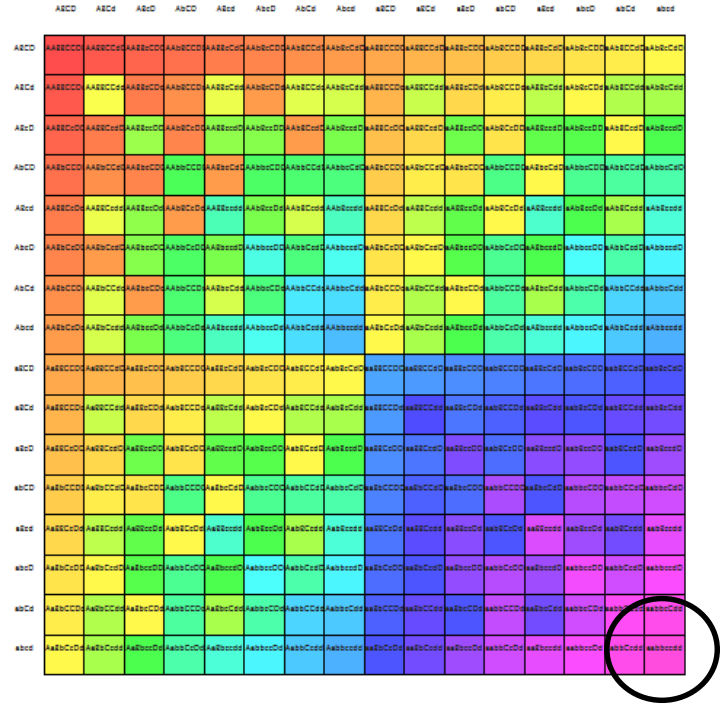
	AB	Ab	aB	ab
AB	AABB	AAbB	aABB	aAbB
Ab	AABb	AAbb	aABb	aAbb
aB	AaBB	AabB	aaBB	aabB
ab	AaBb	Aabb	aaBb	aabb

Total 32 combinations

$2 \times 4^{n/2}$

Non-grafted, 4 genes

Cross:
AaBbCcDd × AaBbCcDd



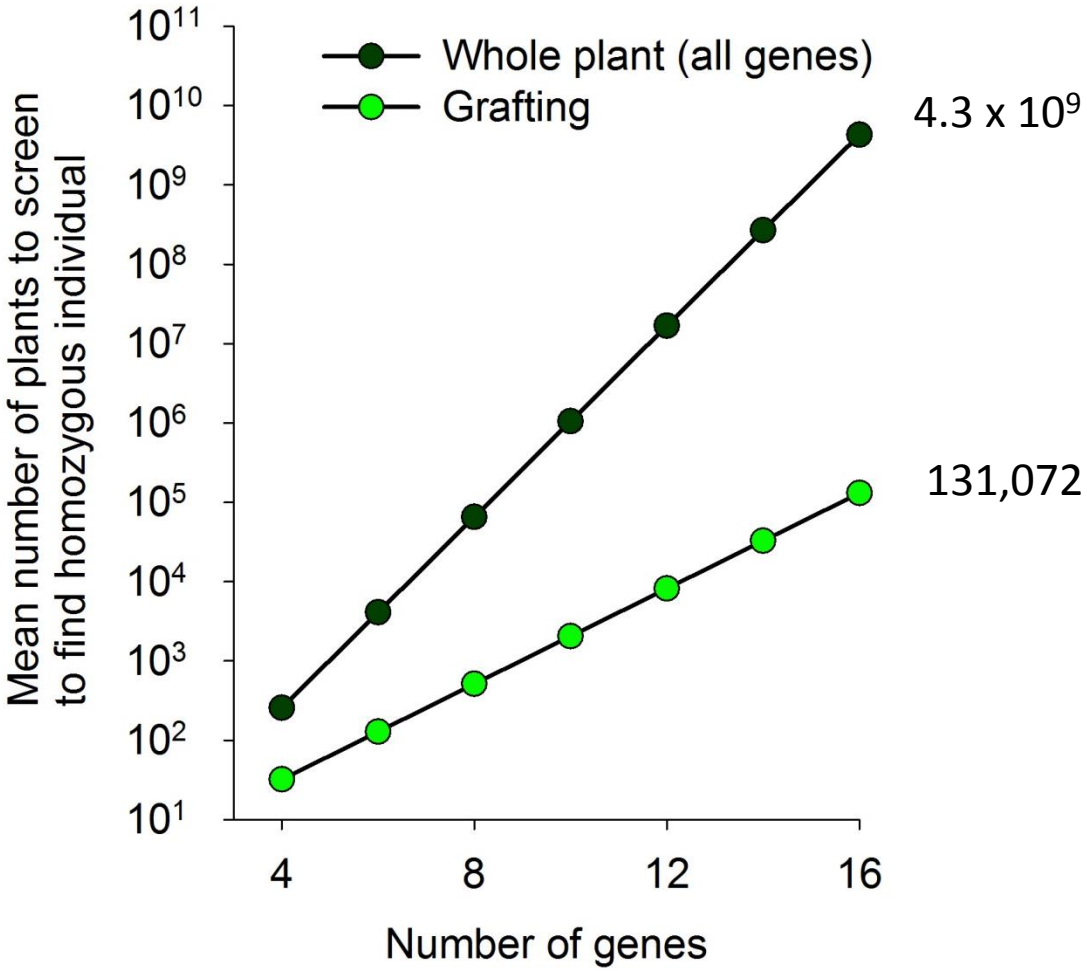
1 in 256 homozygous

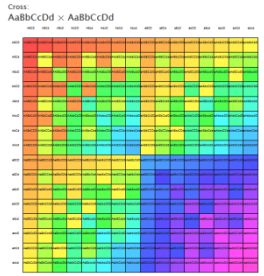
Total 256 combinations

4^{n}

n = number of loci

Mendelian Genetics:
Trait stacking – grafting vs non-grafting



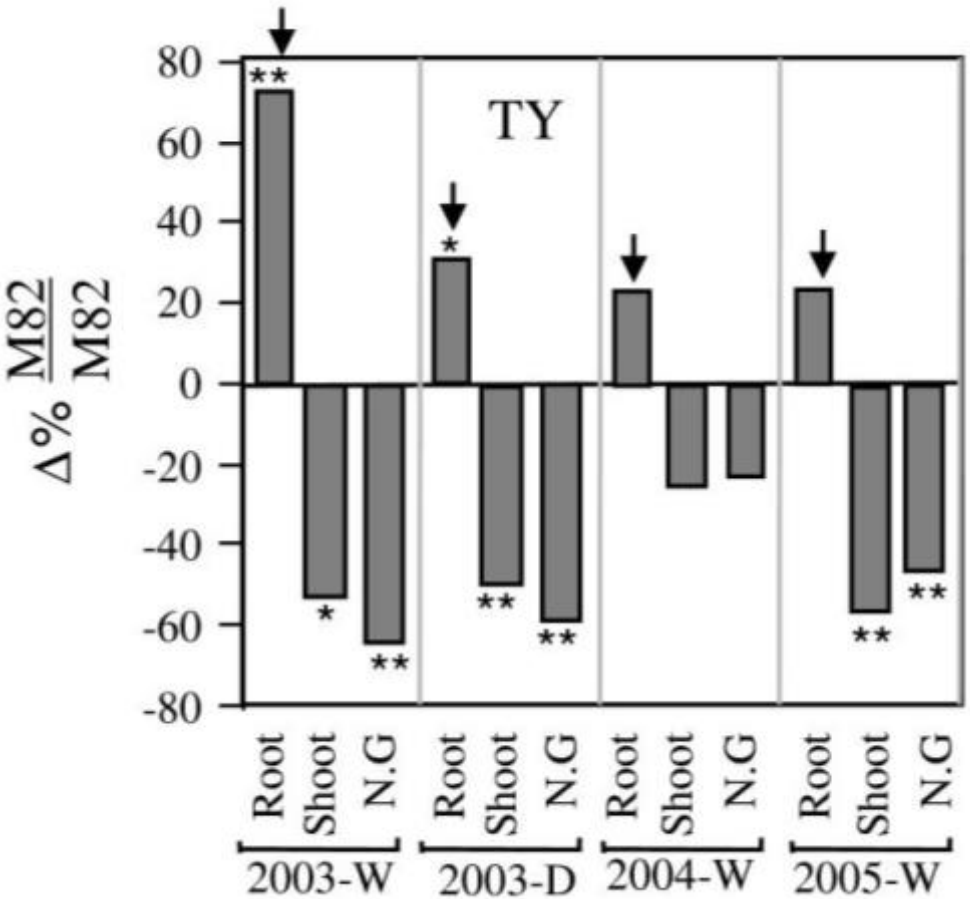


- Assumes equal split of selected genes between scion and rootstock
- Linkage makes the problem much worse

Grafting can overcome pleiotropic effects

IL8-3 (S. pennellii single introgression line)

- IL8-3/M83 give 60% **less** yield than M82/M82
- M82/IL8-3 has 20-60% **more** yield than M82/M82.
- Non-grafted IL8-3 plants do worse than M82/M82

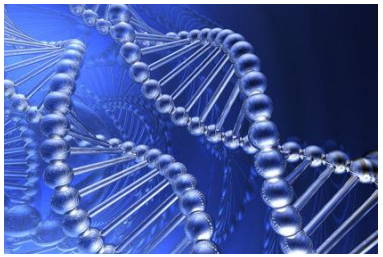


Benefits of IL8-3 can only be achieved when deployed as a rootstock

Gur et al 2011

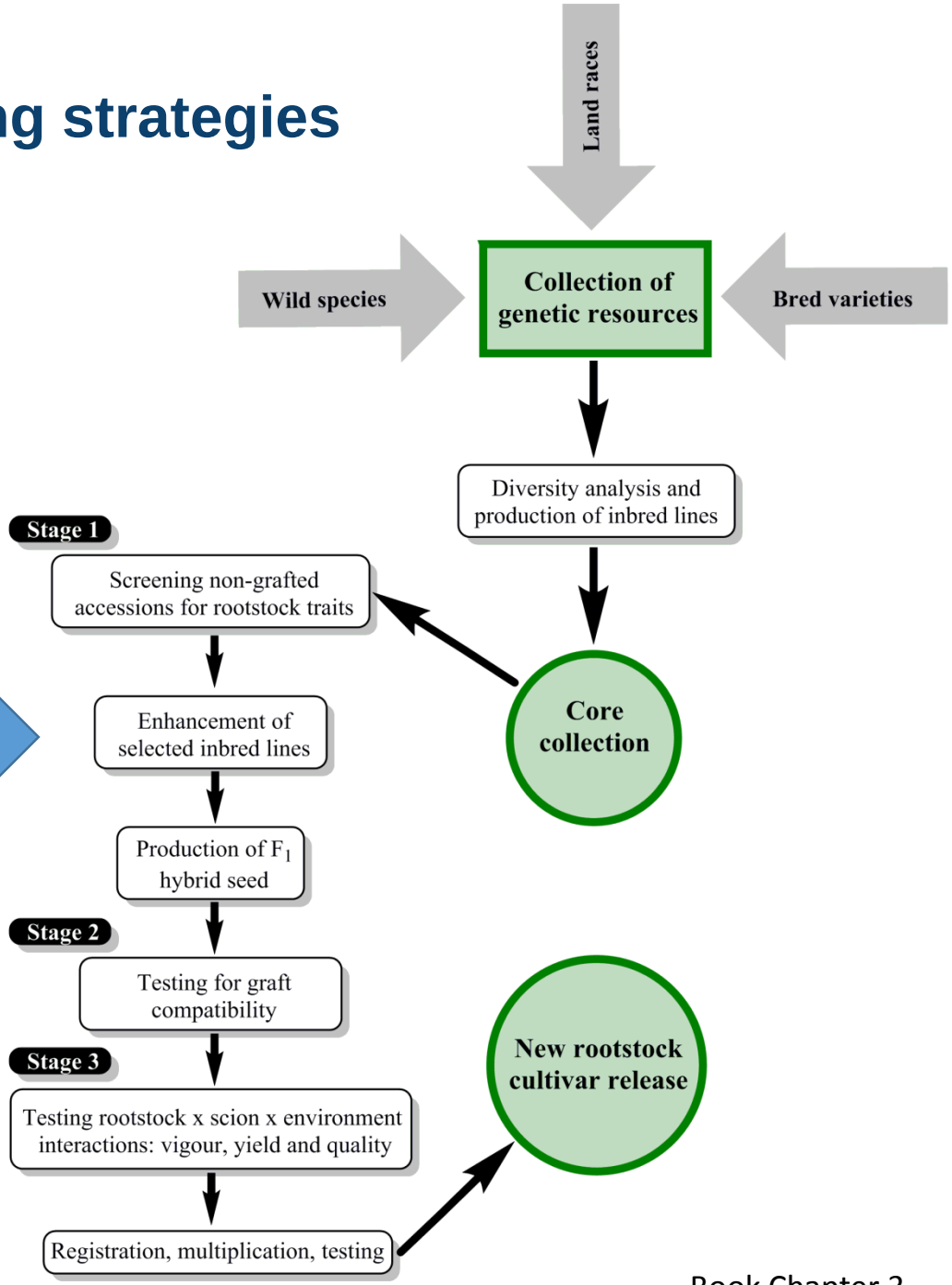
Rootstock breeding strategies

Genomics, mol. genetics



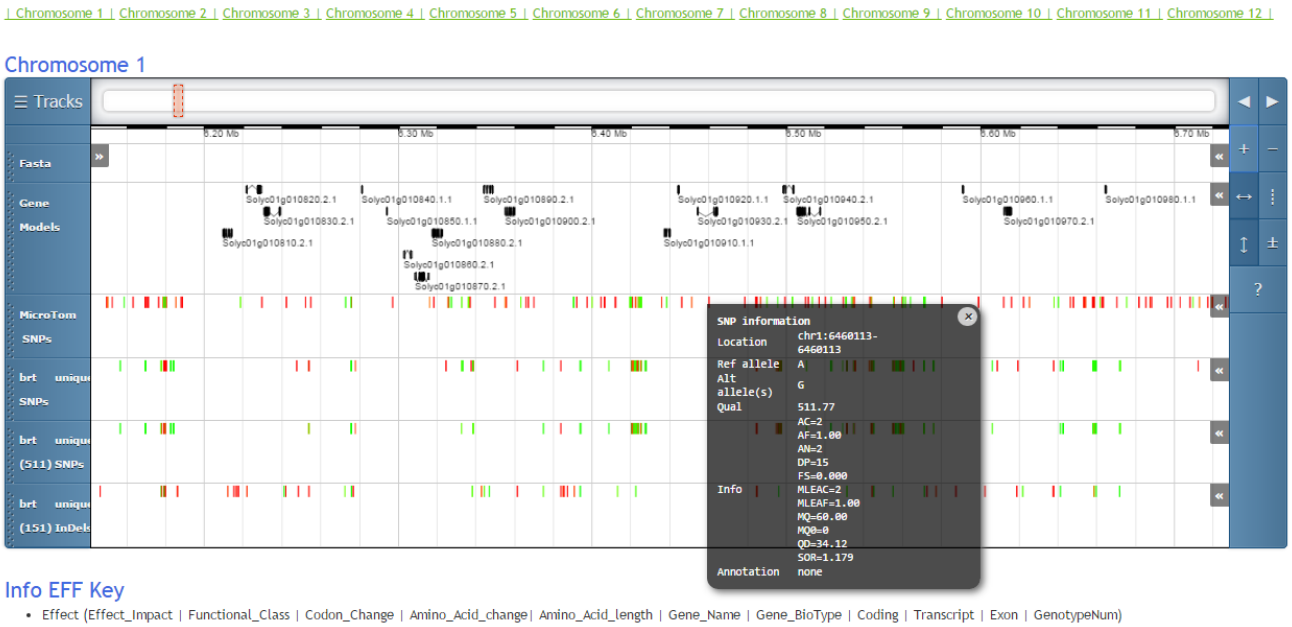
Marker discovery and validation

Marker assisted selection
Genome Editing



Book Chapter 3

Post-genomic era: e.g. look up DNA sequence variation in 500 varieties of tomato on “genome browser”



Third generation sequence technologies are accelerating the pace, e.g.

- Hugely improved pepper genome using “10x Chromium technology”
- SL3.0 tomato reference genome to be released soon, PacBio based reference on its way.
- Understanding biological function limits progress, not genomics

(see WP2, 3 and 4!)

Example 1: exploiting germplasm

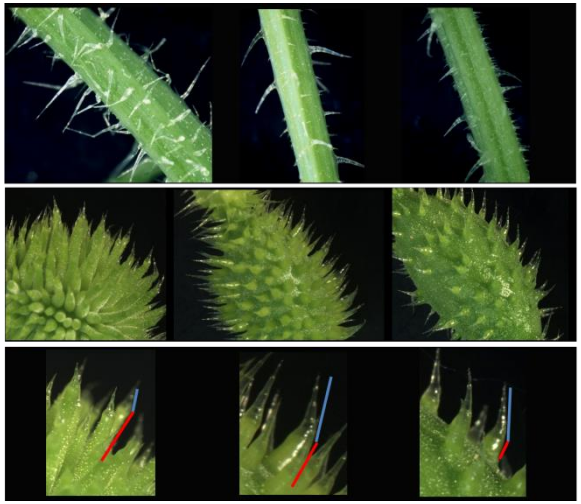
Investigation of wild *Cucumis* species as cucurbit rootstocks to replace standard *Cucurbita maxima* × *Cucurbita moschata* F_1 hybrids

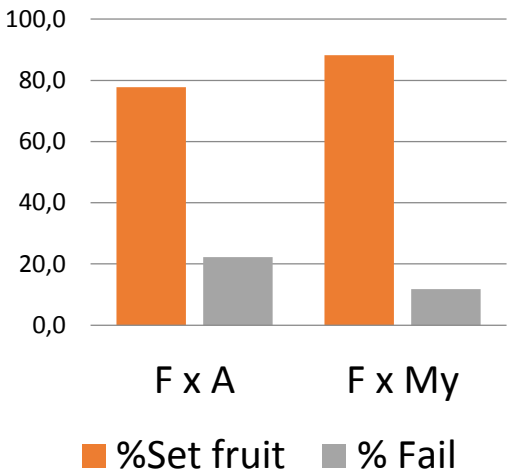
Carmina Gisbert and Belen Picó

Parents tested as hybrids grafted to melon:

C. metuliferus (M),
C. ficifolius (F),
C. myriocarpus (My),
C. Zeyhery
C. anguria (A)

F *A* *F* x *A*





Cross-compatible

F x *A* and *F* x *My* selected as:

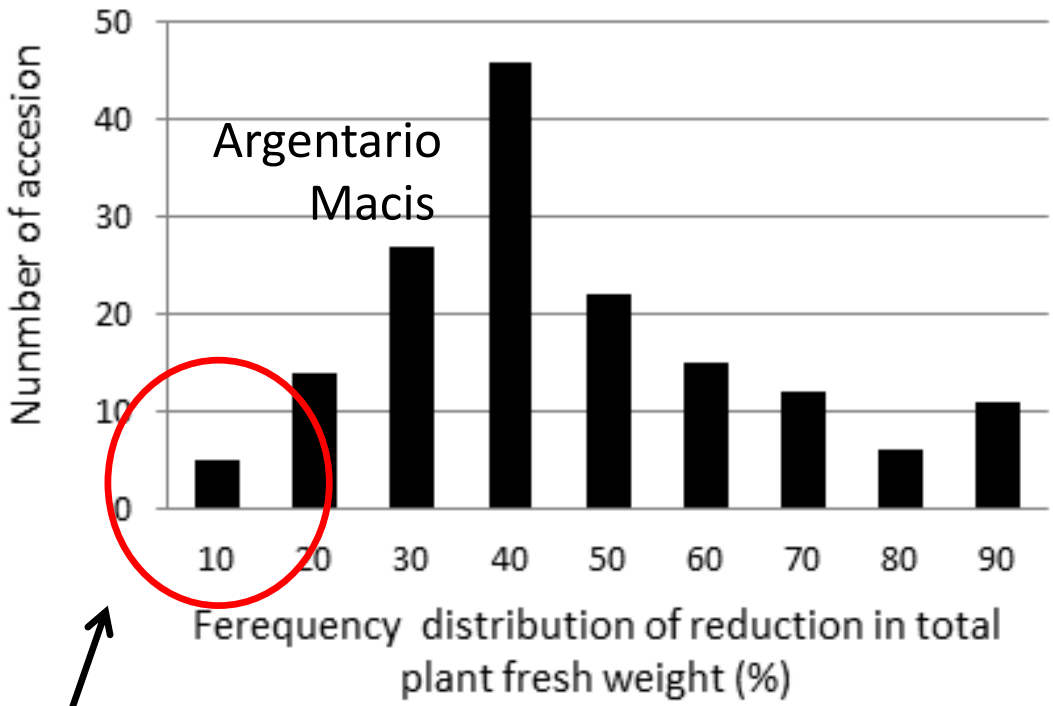
- Cross-compatible
- Good graft compatibility with melon
- 100% *Fusarium* wilt resistance

Fusarium wilt resistance

Example 2: exploiting germplasm

Screening Turkish Bottle Gourd (*Lagenaria siceraria*), germplasm for salinity resistance

Halit Yetişir et al



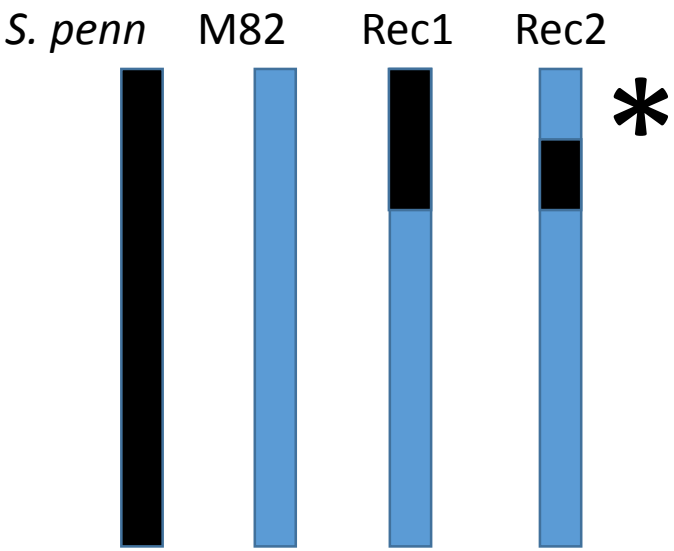
160 accessions were screened at 10 dS m^{-1} salinity in a hydroponic system (n = 3). Positions of two commercial rootstocks are given

A significant number of accessions showed less growth reduction than the commercial rootstocks

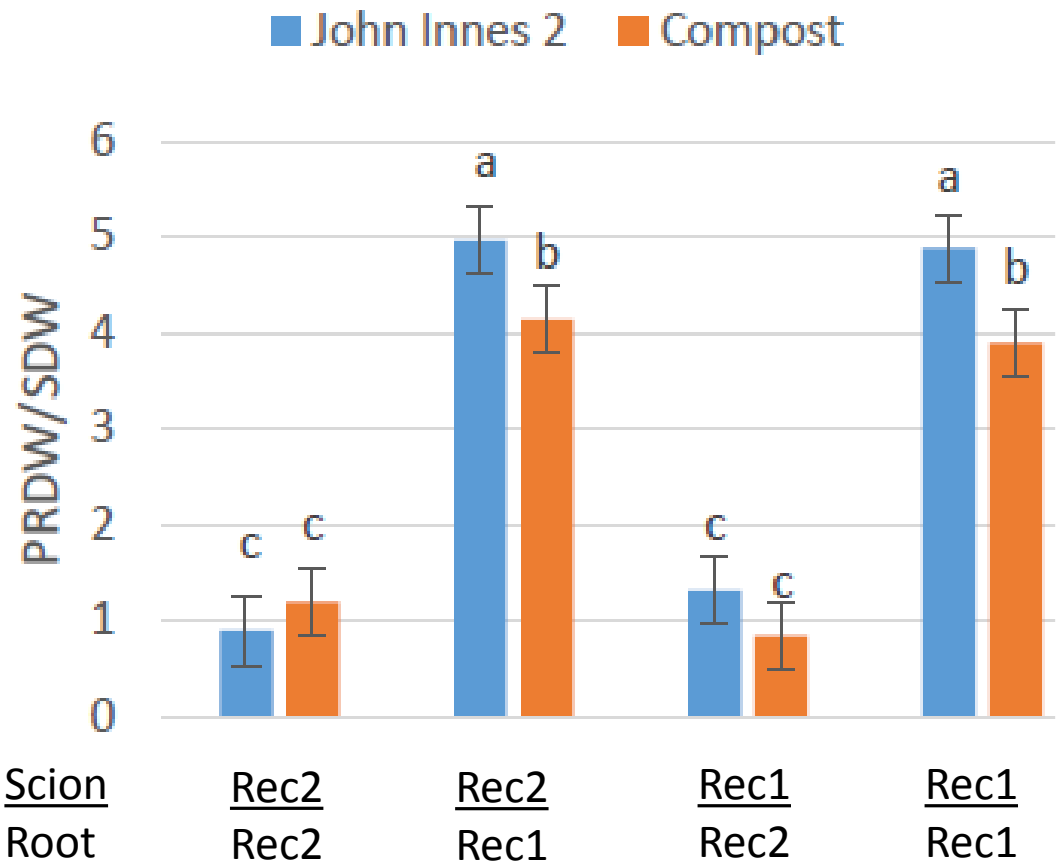
Example 3: searching for molecular markers

Fine mapping of a QTL for root vigour in tomato to an interval of eight genes (*S. lyc.* × *S. pennellii* introgression lines)

Chromosomes:
parents and recombinants



Root vigour in reciprocal grafting



N = 6, error bars = LSD 5%

Thompson et al

Conclusions

Thanks!

- WG1 has been beneficial for collaboration including with seed companies
- Book chapters 2 and 3 will be useful resources.
- Genomic technologies are running way ahead of biological understanding
- Public research and is vital to increase understanding of traits and to disseminate information on useful germplasm