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Precision breeding for complex traits using NBTs - from research to applications

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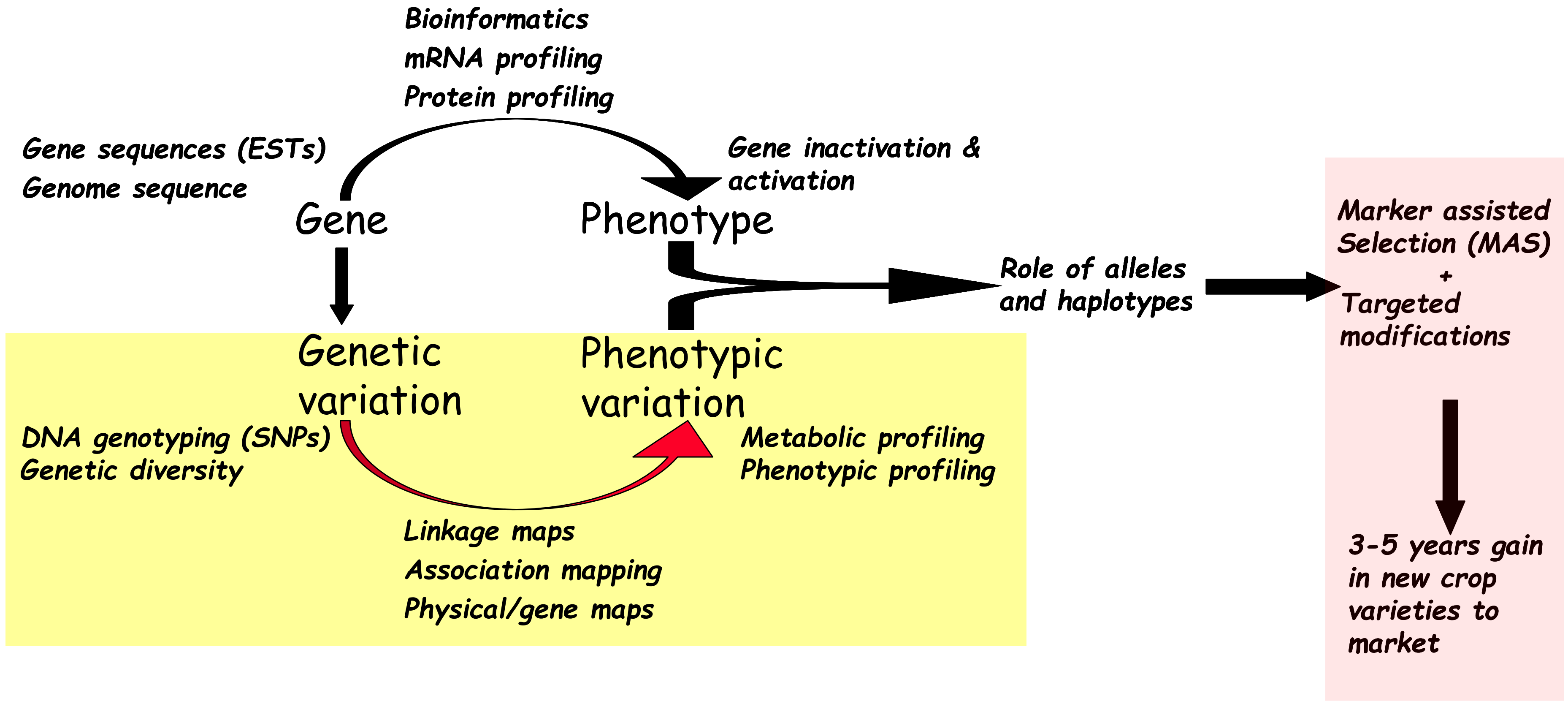
IGA
ISTITUTO DI GENOMICA APPLICATA

What are the current limitations to the use of GE for complex traits

- Scientific:
  - Knowledge about genes
  - Knowledge about gene regulation
  - Knowledge about networks
- Regulatory:
  - Regulation of genome editing in Europe currently in flux: perspectives
- Societal
  - How to win consumer acceptance of GE

BREEDING MEETS GENES THROUGH GENOMICS: LINKING GENETIC AND PHENOTYPIC VARIATION



Bioinformatics
mRNA profiling
Protein profiling

Gene sequences (ESTs)
Genome sequence

Gene inactivation & activation

Gene

Phenotype

Role of alleles and haplotypes

Marker assisted Selection (MAS)
+
Targeted modifications

Genetic variation

Phenotypic variation

DNA genotyping (SNPs)
Genetic diversity

Metabolic profiling
Phenotypic profiling

Linkage maps
Association mapping
Physical/gene maps

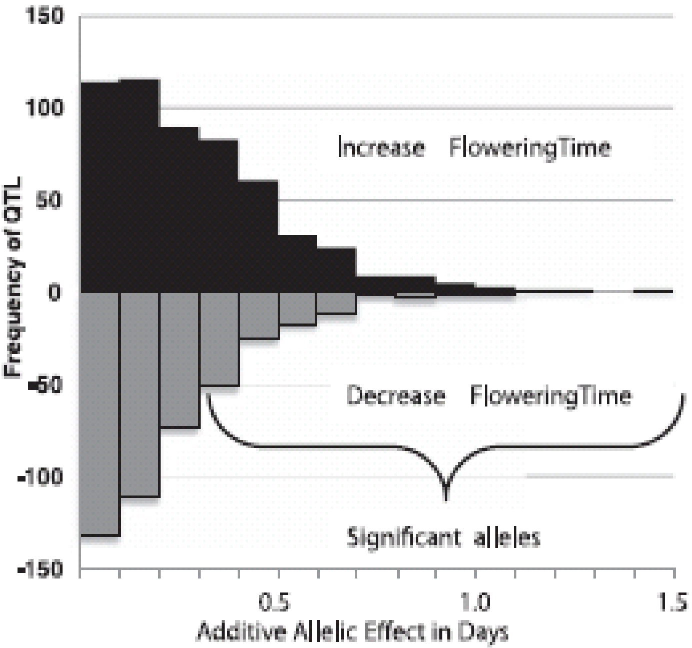
3-5 years gain in new crop varieties to market

Modified from Morgante and Salamini, 2003

PRECISION BREEDING

- It is possible to identify genes that control a range of agronomically relevant traits
  - Technological and scientific developments are accelerating this process: there is a need for investments and research
  - Knowledge-based breeding
- Targeted modifications of genes
  - New variability is created
  - *In planta* mutagenesis aimed at obtaining the desired mutations: genome editing
  - Cisgenic approach
  - Transgenic approach???

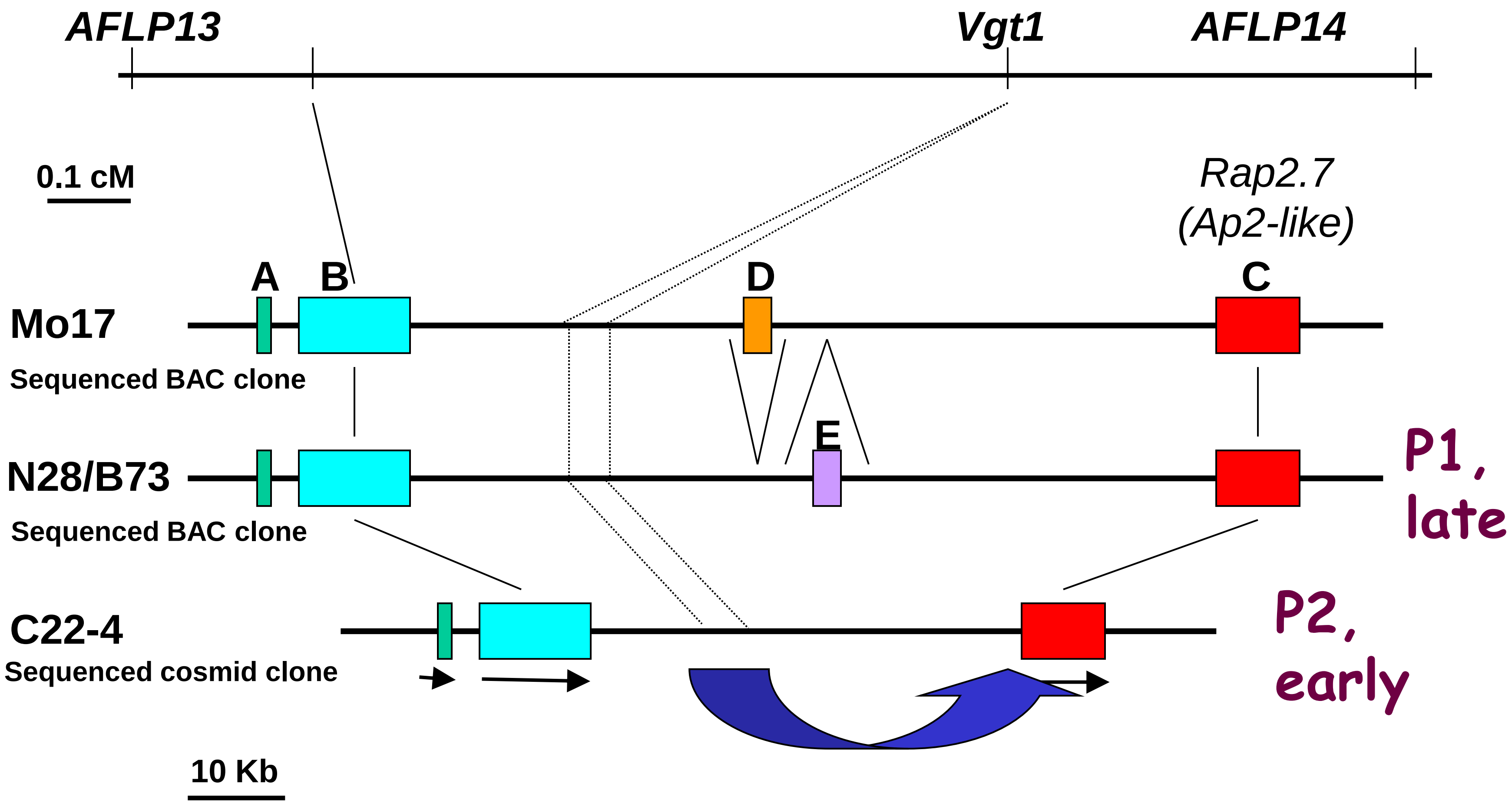
MANY QTL LOCI AFFECT A SINGLE TRAIT



- 39 QTLs affect days to silking (flowering time) in maize
- No large effect QTL
- Only one has been cloned: Vgt1

Buckler et al., Science, 2009

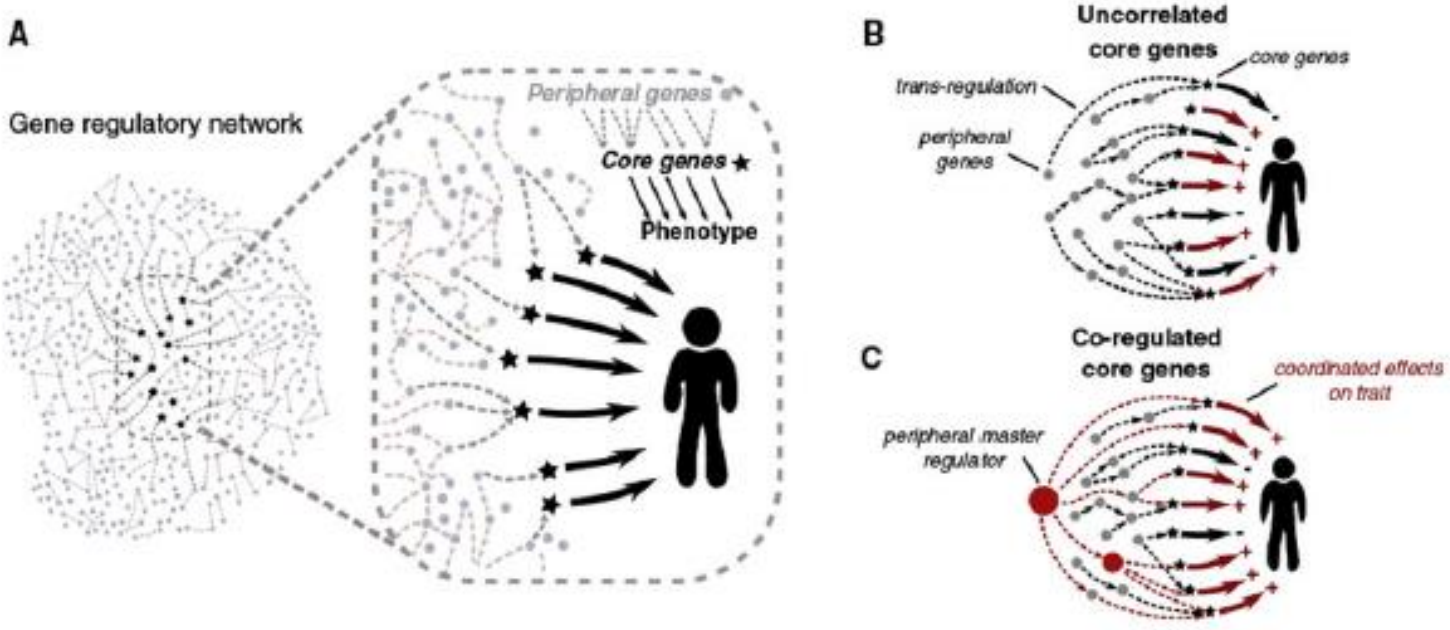
POSITIONAL CLONING OF VGT1, A CIS-REGULATORY REGION



Salvi et al., PNAS 2007

An Expanded View of Complex Traits: From Polygenic to Omnigenic

Evan A. Boyle,[1,*] Yang I. Li,[1,*] and Jonathan K. Pritchard[1,2,3,*]
Cell *169*, June 15, 2017

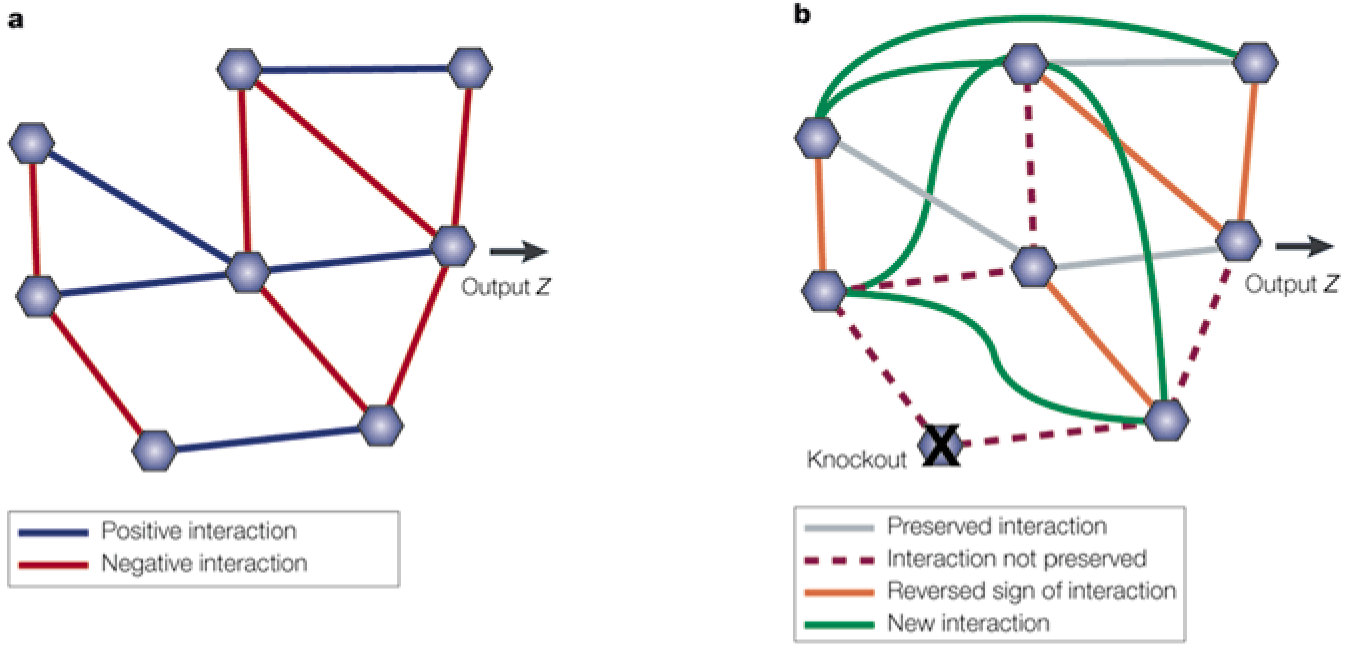


Trans **Effects on Gene Expression Can Drive Omnigenic Inheritance**
2019, Cell *177*, 1022–1034

Xuanyao Liu, Yang I. Li,
Jonathan K. Pritchard

NETWORK DEGENERACY AND REDUNDANCY

Degeneracy in biological systems



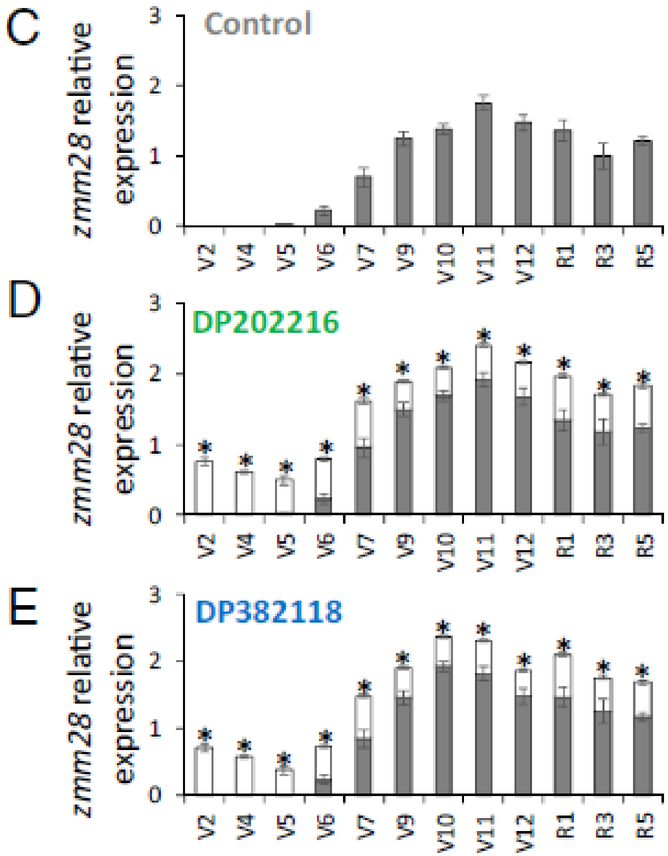
Interactions in (**a**) a hypothetical gene network, and in (**b**) the same network with one gene knocked out. When one element of the system is changed ('knocked out'), the rest of the system changes in response. In this instance, the output (*Z*) is unchanged, illustrating the phenomenon of degeneracy. Under different conditions, the output might be different, producing a mutant phenotype in some cases, or a new emergent property in others. 'Positive' and 'negative' refer to whether a phenotype improves or degrades as a result of the interaction. 'Reversed sign' refers to a change in the direction of effect in that interaction.

Greenspan RJ, *Nature Reviews Genetics* **2**; 383-387 (2001)

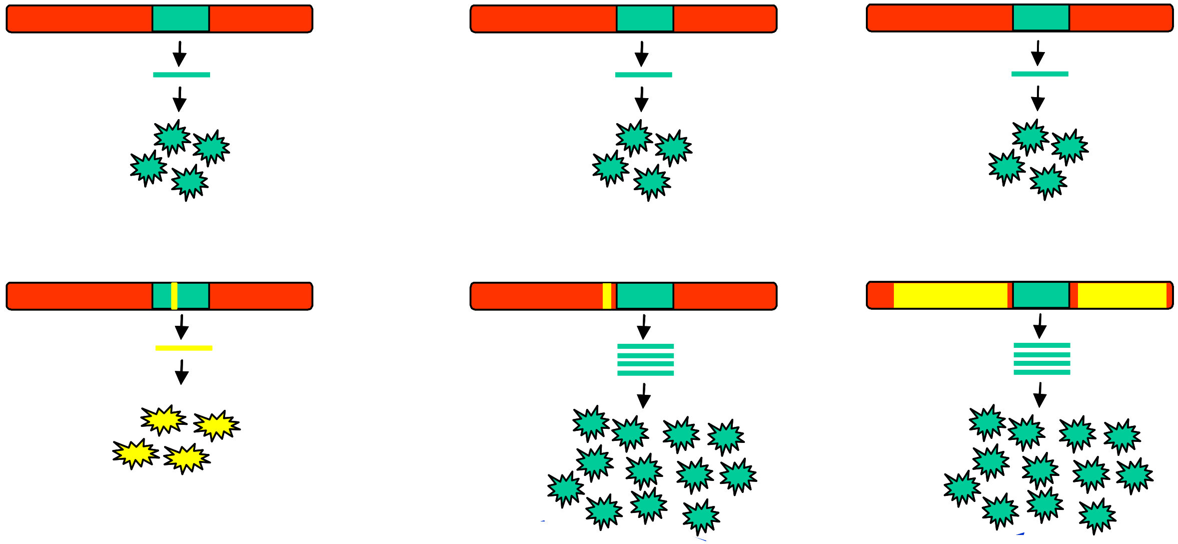
GENETIC MODIFICATIONS FOR YIELD IMPROVEMENT

Overexpression of *zmm28* increases maize grain yield in the field

Wu et al. 2019 PNAS 116, 23850-23858



SEQUENCE VARIATION AND FUNCTIONAL POLYMORPHISMS



Coding variation

Cis-Regulatory variation

Humans, mouse

Plants

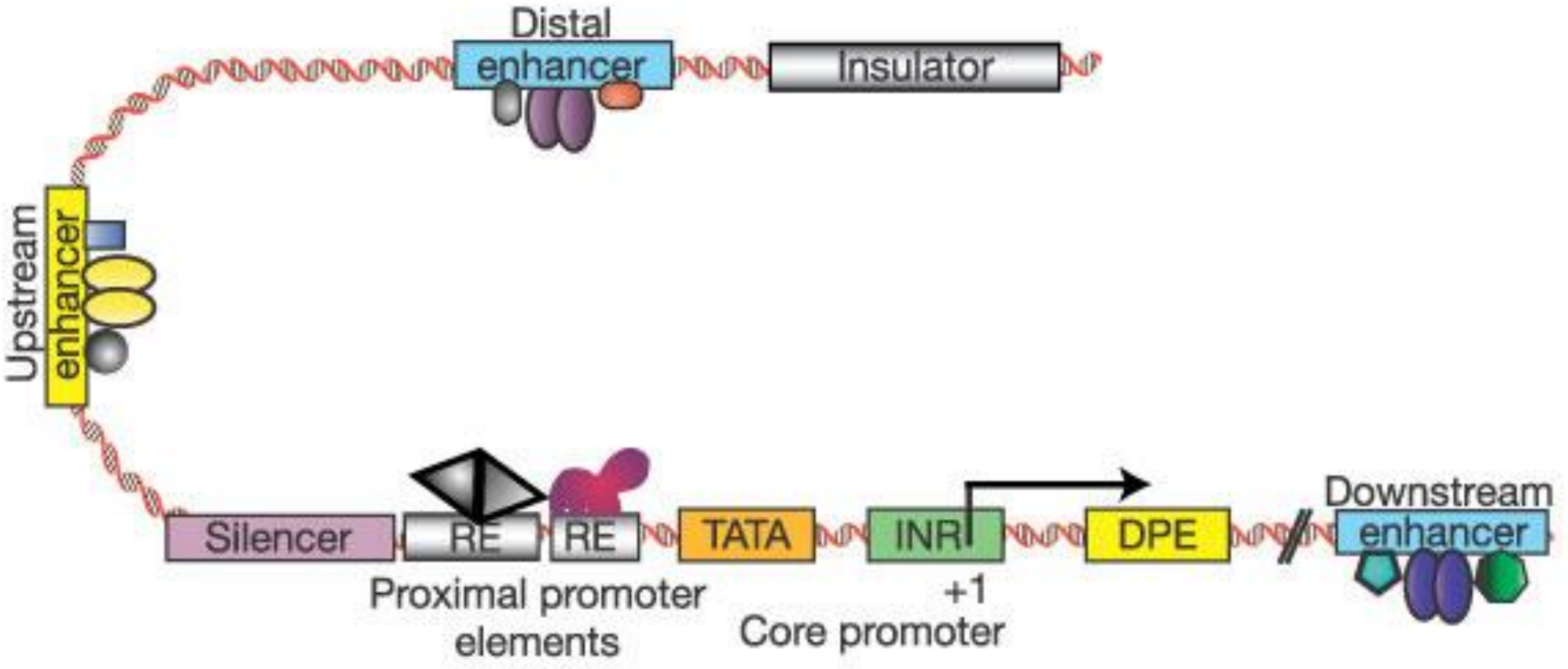
Allele A

Allele B

Allele A

Allele C

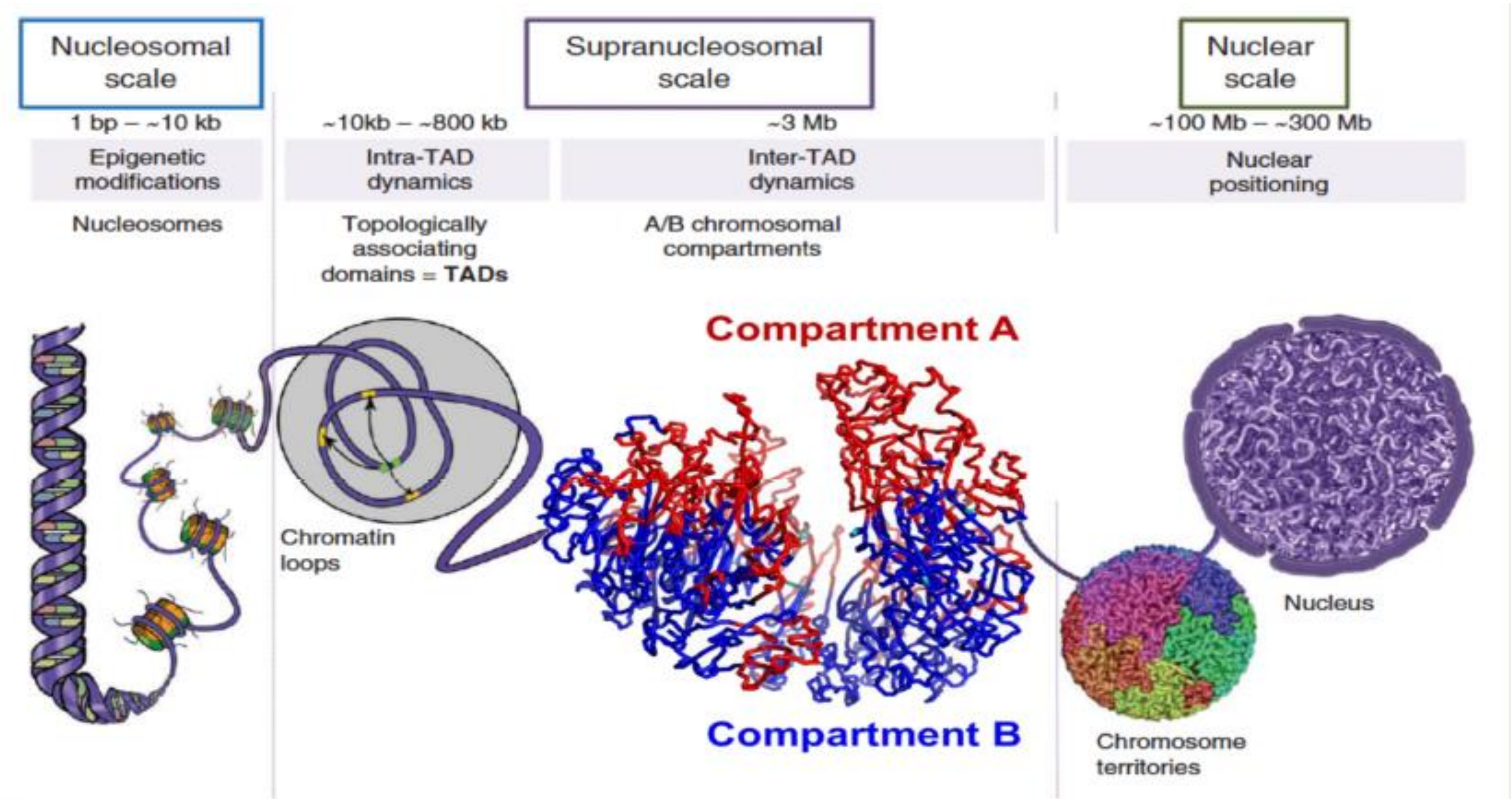
CIS-ACTING TRANSCRIPTIONAL CONTROL: A COMPLEX MODULAR SYSTEM



Levine & Tjian, Nature, 2003

Long range effects are frequently observed

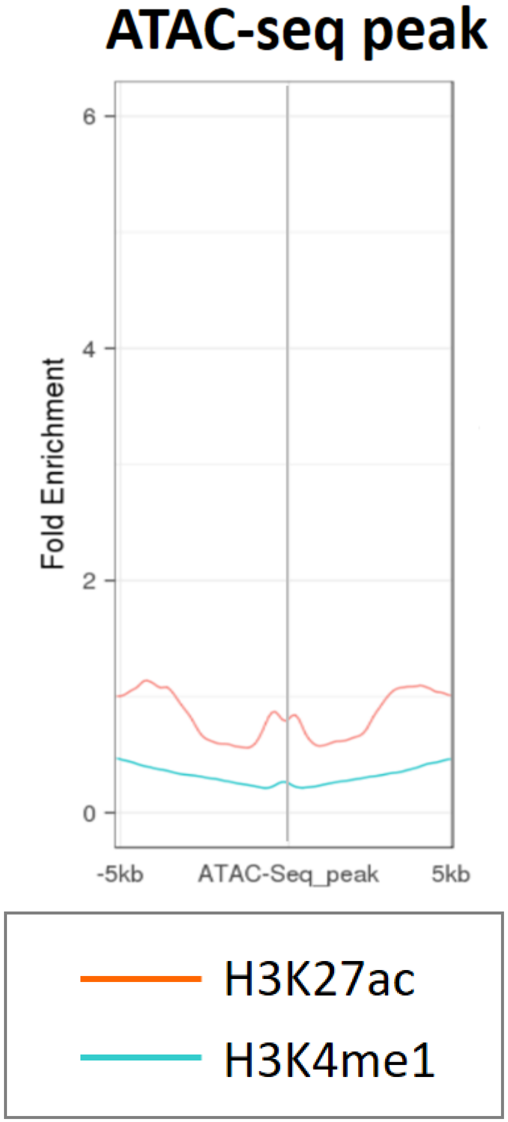
CIS-ACTING TRANSCRIPTIONAL CONTROL: A 3-D VIEW

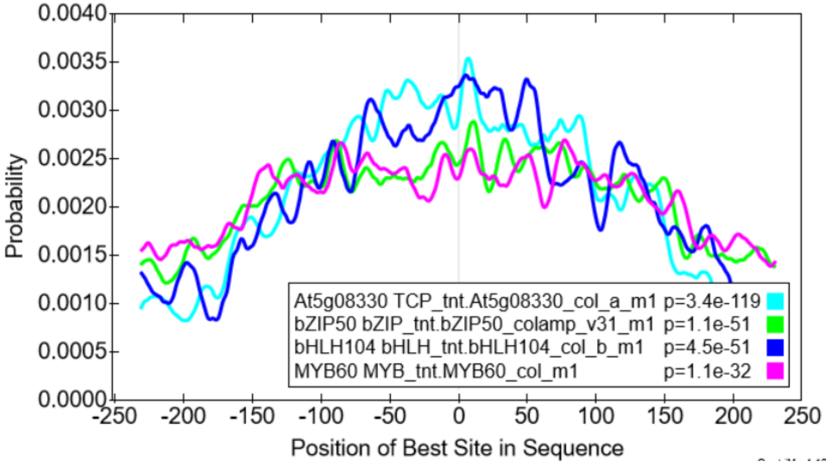


Adapted from Nature, 2017

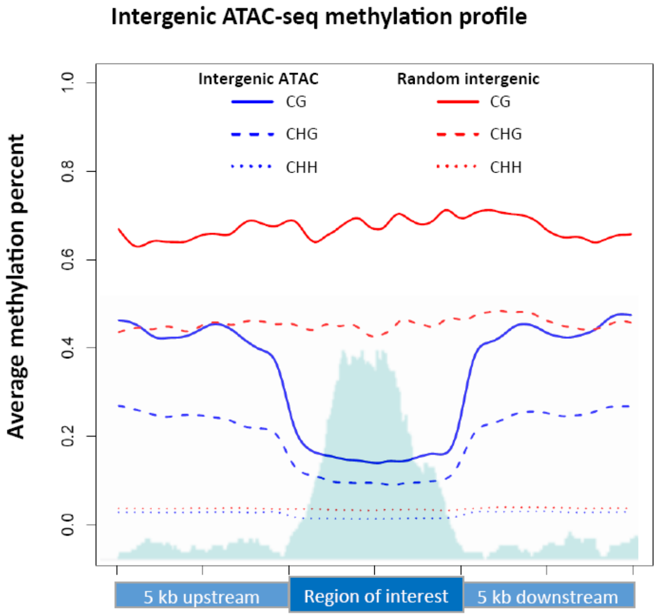
Role of liquid-liquid phase separation phenomena vs. diffusion kinetics?

Intergenic open chromatin regions lack expected histone modifications but are enriched in TFBS and hypomethylated



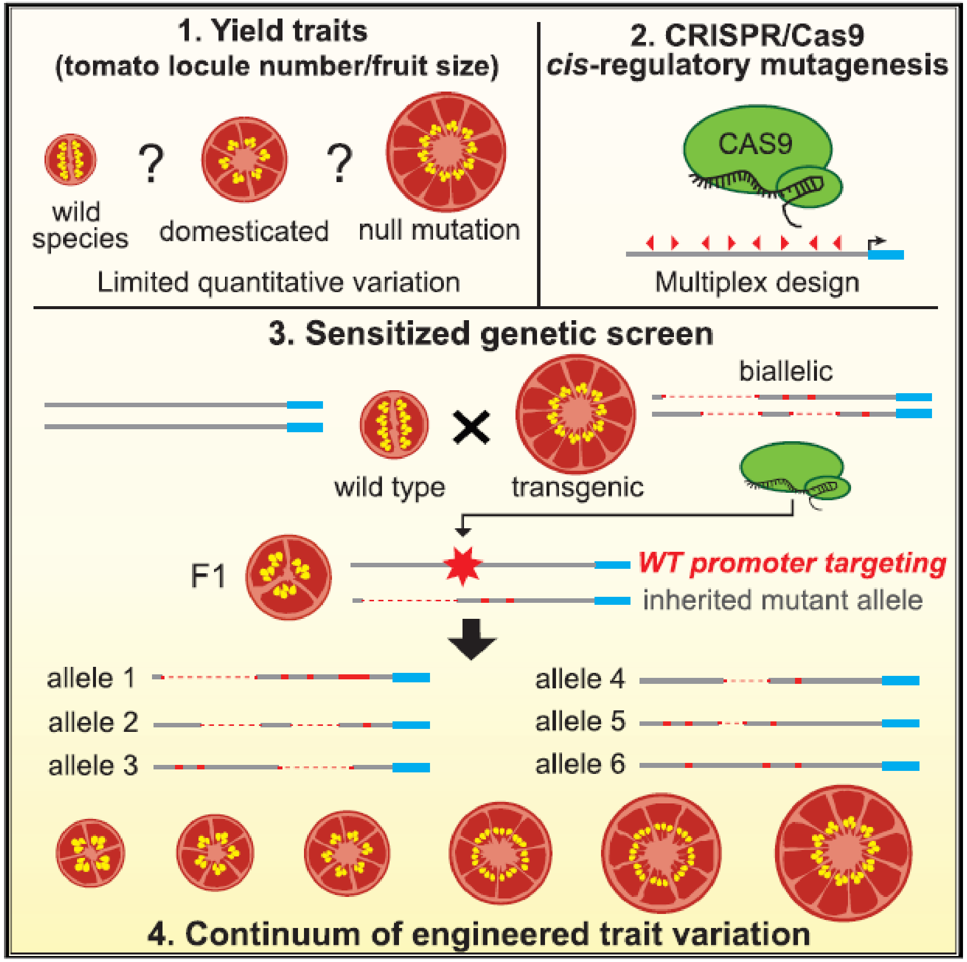


Motifs are centrally enriched in intergenic ATAC-seq peaks



Schwope et al. Plant J., in press

EDITING CIS-REGULATORY VARIATION



Engineering Quantitative Trait Variation for Crop Improvement by Genome Editing

Rodriguez-Leal et al. 2017 Cell 171, 470-480

INGREDIENTS NEEDED TO DEPLOY INNOVATIONS IN AGRICULTURE

(*i.e.* how to go from laboratory to tables)

1. Excellent research
2. An innovation ecosystem
3. An appropriate regulatory framework
4. Consumer acceptance

NBTs:
REGULATORY HURDLES

- Varieties from NBTs
  - – Cisgenesis treated as transgenesis
    - Despite EFSA 2012 scientific opinion
  - – Genome editing treated as transgenesis
    - ECJ sentence July 25th, 2018
    - Complex and illogical situation
    - Undistinguishable and not traceable
  - – New hopes from recent EU Commission study

HOW TO CHANGE PERCEPTION OF INNOVATION IN AGRICULTURE

- New Breeding Technologies together with digital agriculture at the center of a revolution in farming
- Combine productivity and sustainability
- Innovations to preserve traditions
- Innovations to preserve agricultural and food diversification

THE FUTURE OF REGULATIONS FOR GENETIC MODIFICATIONS

- Future decisions should descend from logical and not ideological considerations
- Consider all implications and consequences of regulation
  - On consumers
  - On farmers
  - On world trade
  - On environment