SOME EQUIPMENTS...

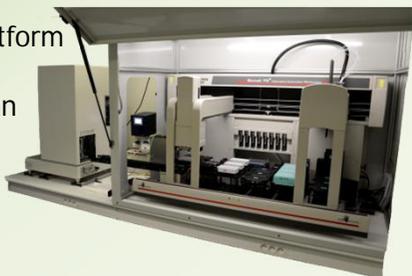**DNA extraction platform**

Biomek Fx, Beckman

Droplet Digital PCR system

Qx200TM AutoDGTM ddPCR, Bio-Rad

Illumina sequencers

NextSeq500   MiSeq

Plant growing facilities

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ACCESS

EPITRANS is part of the Institute of Plant Sciences of Paris-Saclay (IPS2)

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EPITRANS

Exploring and creating genetic and epigenetic diversity on crops

Agricultural yields have been greatly enhanced in the past 50 years. However, climatic perturbations and the spread of pests and pathogens are major upcoming threats to agriculture worldwide. Crop selection led to a reduction in the allelic variability and thereby narrowed the possibilities for genetic improvement.

In order to carry on the genetic improvement process, we need to identify new phenotypes (i), identify the genetic basis underlying those phenotypes (ii) and finally transfer those phenotypes in crops (iii).
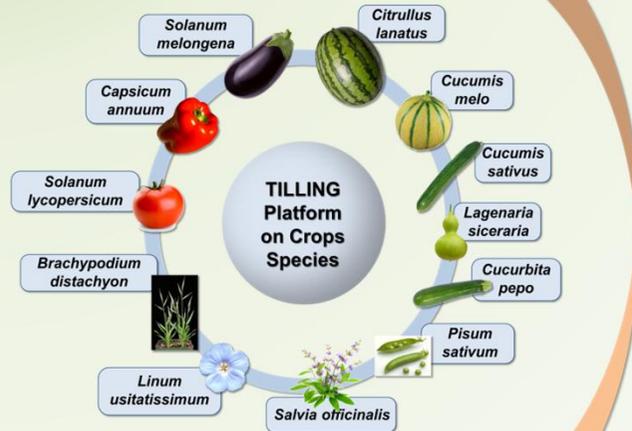
Thanks to last decades'efforts in plant sciences, many of the genetic and epigenetic regulators controlling key aspects of plants developments and plants interactions with their environments are known: stress resistance, photosynthesis, defense against pathogens etc. Despite this massive amount of data, the translation of the findings into traits in elite varieties is modest.
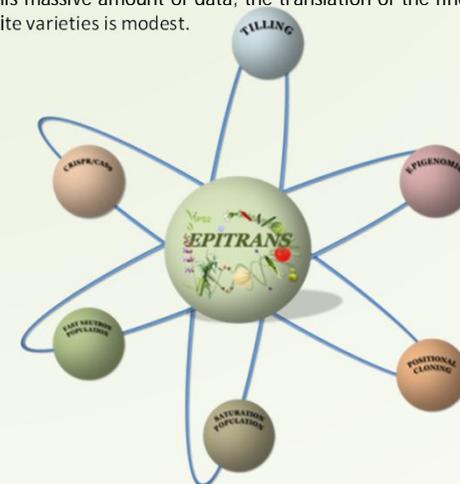
To help to translate fundamental research into leader alleles EPITRANS platform raise the challenge of helping the scientific community to investigate agronomic traits in model and crop species.

TILLING : Alleles Engineering

EPITRANS — Epigenomic & Translational Research on Crops

POPULATIONS CREATION

TILLING Platform on Crops Species

- Solanum melongena
- Citrullus lanatus
- Capsicum annuum
- Cucumis melo
- Cucumis sativus
- Solanum lycopersicum
- Lagenaria siceraria
- Brachypodium distachyon
- Cucurbita pepo
- Pisum sativum
- Linum usitatissimum
- Salvia officinalis

PCR-based indexed libraries

NGS-SCREENING

BIO-INFORMATIC ANALYSIS

SENTINEL application

ALLELES STACKING & PLANTS PROTOTYPES

Epigenomic Mark

EPITRANS — Epigenomic & Translational Research on Crops

- DNA methylation: MeDIP, Bis-seq
- Chromatin Isolation Through RNA Purification: ChIRP
- Chromatin Accessibility: ATAC-seq
- 3D chromatin Architecture: ChIAPET, Hi-C
- TF-ChIPseq
- Histone modifs: ChIPseq
- Nucleosome positioning: Mnase-seq

5-mC, 5-hmC

H3K4me1, H3K4me2, H3K4me3, H3K27ac
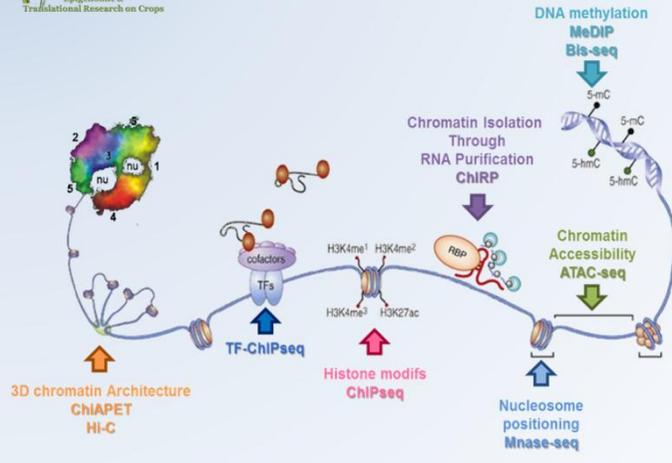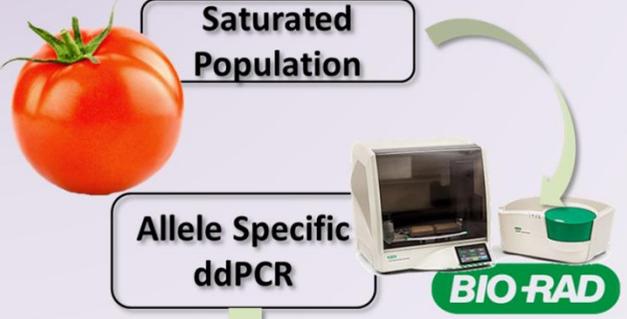
Many studies have highlighted the determining role of epigenetic regulations, not only in plant adaptation to stress and changing environments but also in the control of agronomically important traits. A better understanding of such processes provides the tools necessary to the development of innovative and efficient approaches to crop improvement.
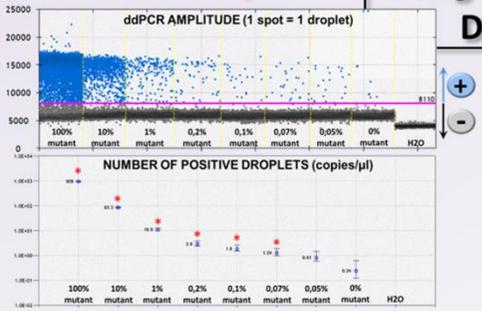

Positional Cloning
Bulk-Segregant Analysis Approach

Parental Lines

WT × Mutant

Phenotyping

F1

F2

Candidate Genes

CLCbio

Causal variant identification

Bulk sequencing

Saturation Mutagenesis

EPITRANS — Epigenomic & Translational Research on Crops
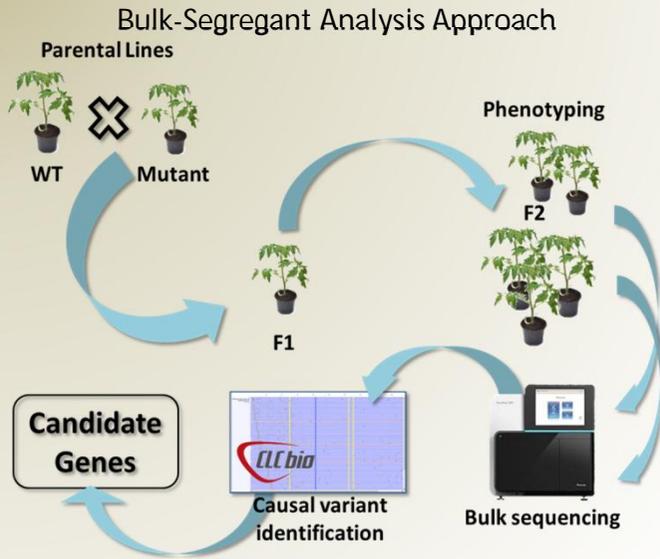
Set up a population of mutants which carrying up to 90% of the putative EMS mutation

Saturated Population

Allele Specific ddPCR

BIO-RAD

Targeted SNP Detection

ddPCR AMPLITUDE (1 spot = 1 droplet)

100% mutant, 10% mutant, 1% mutant, 0,2% mutant, 0,1% mutant, 0,05% mutant, 0% mutant, H2O

NUMBER OF POSITIVE DROPLETS (copies/µl)

100% mutant, 10% mutant, 1% mutant, 0,2% mutant, 0,1% mutant, 0,07% mutant, 0,05% mutant, 0% mutant, H2O

On development...

Fast Neutron mutagenesis

First FN population on tomato

New Screening methods

- SMALL INDEL < 500pb → PCR-based libraries sequencing
- BIG INDEL > 1 kb → ddPCR screens

CRISPR/Cas9 Platform

- Producing on demand mutant of tomato

Exome Sequencing

- Sequencing whole exome of mutant lines
- Targeted only mutation impacted protein sequence
- Generate database of mutations