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High throughput phenotyping... What's going on

Christophe Salon

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High throughput phenotyping..

What's going on

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Goals

Scientific : Understand plant response to environmental factors : get a enhanced agronomic production, with a better quality, less fertilizers...

Technical : Reduce the gap between genotyping and phenotyping flows

Economic : Offer to companies the possibility to SME the possibility to access/develop tools and methods at the same level as foreign ones

Goals

Offer to the plant science community high throughput phenotyping infrastructure :

Academic and private Research, breeding companies, technical institutes:

Platforms + associated methods and tools

Characterize large genotype series such as needed for genetic variability studies:

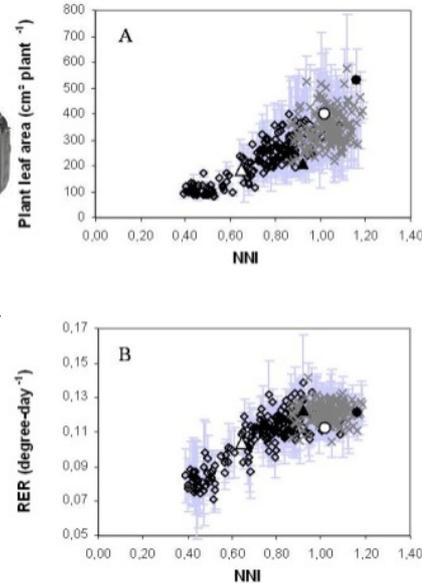
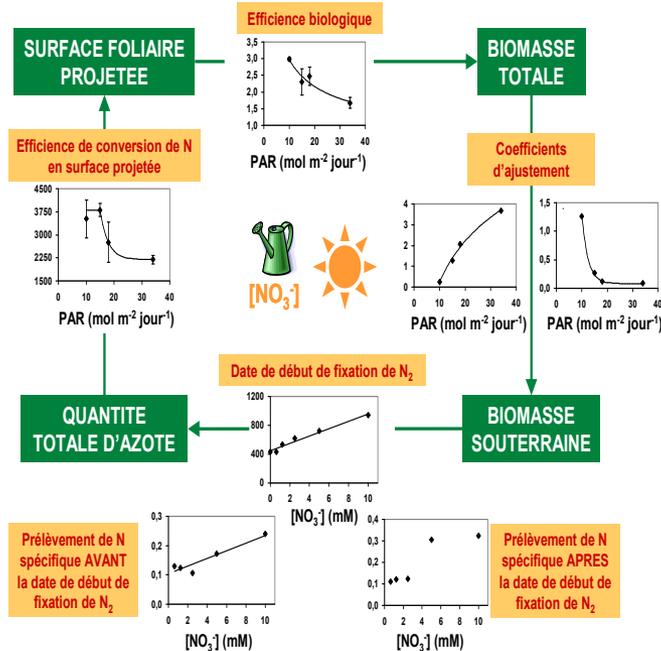
100x field plots- 1000x plants

Populations - varieties- mutants - GMO

Create diverse environmental scenarios including climatic changes

hydric constraints, N stress, parasites, combinations with other constraints : [CO₂], temperature, ...

Before: Detect contrasted N nutritions



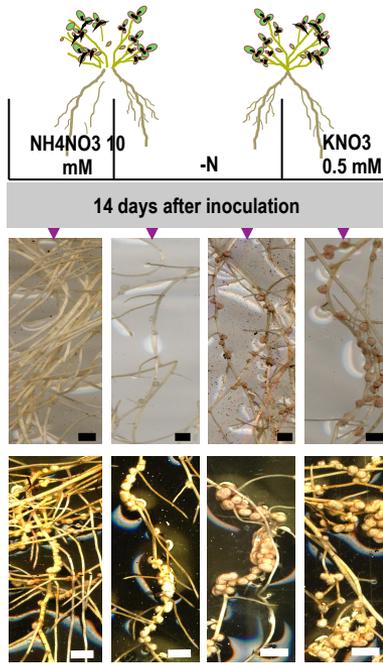
Genotypes of Medicago RIL ranked for ability to uptake N

Moreau et al. (2006), *Plant, Cell and Envir.*, 29:1087-1098.
 Moreau et al. (2007), *Plant, Cell and Envir.*, 30:213-224.
 Moreau et al. (2008), *J. Exp. Bot.*, 59:3509-3522.

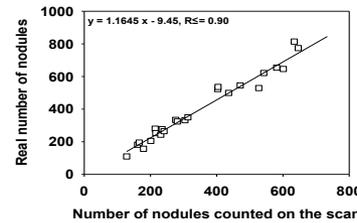
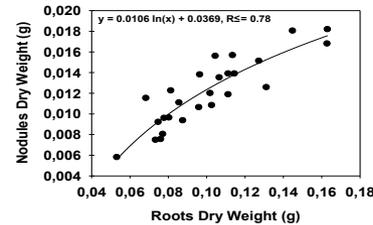
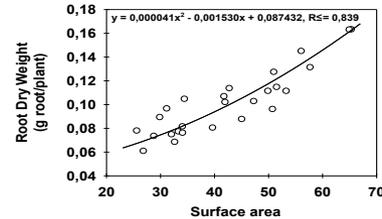
Before: Identify a strategy



Adaptative strategy of plants faced to a N constraint



Split roots



**Morphometry
versus
functional
strategy
identification**

**Nodules number and
size, appearance**

Ruffel et al. (2008), *Plant Physiol.* 146: 2020-2035.

Salon et al. (2009), *CRAS*, 332 :1022-1033.

Jeudy et al. (2010), *New Phytol.*, 185:817-828.

Before: Functional characterization of a mutant

Characterizing the functional behavior of a mutant: **hyper ramified TR 185**

Wild Type

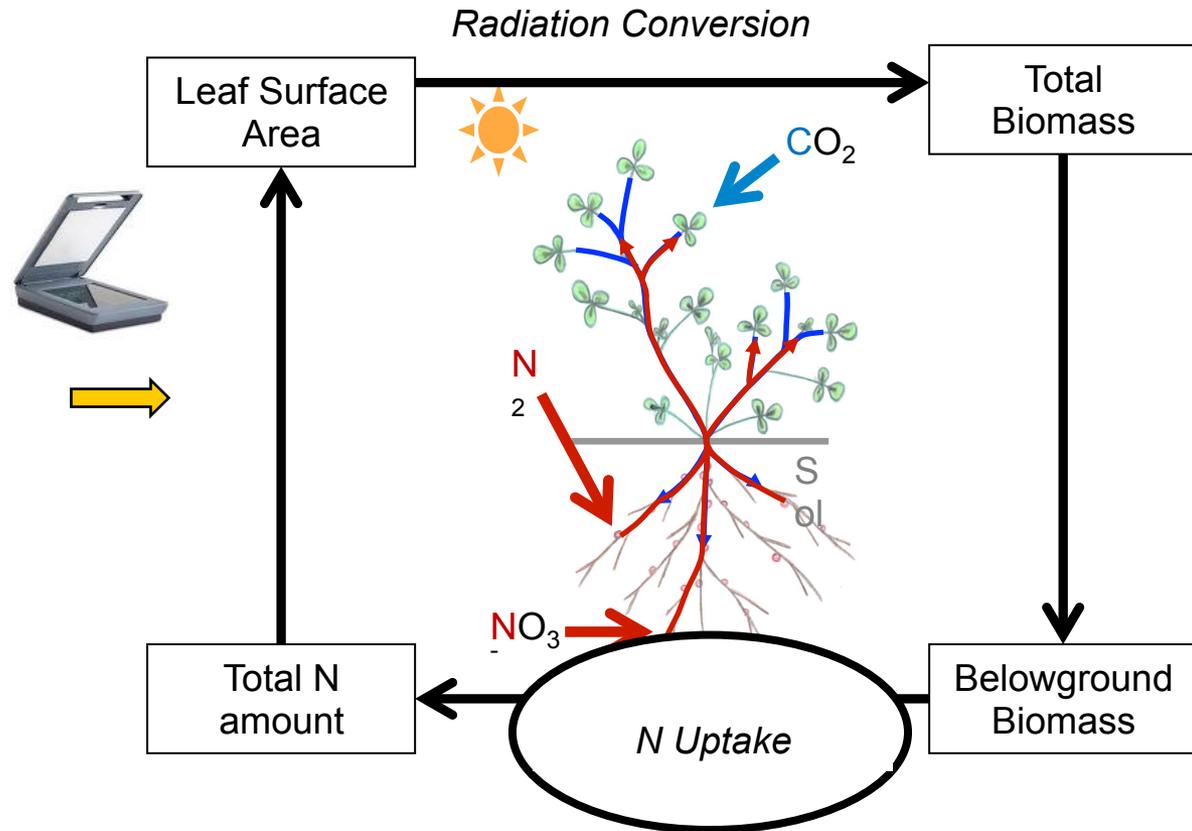
Mutant = TR185



Contrasted root architecture



No modifications of C acquisition rules
Lower N uptake efficiency



Salon et al. CR Biologies 2009

Bourion et al submitted

Food for thoughts...



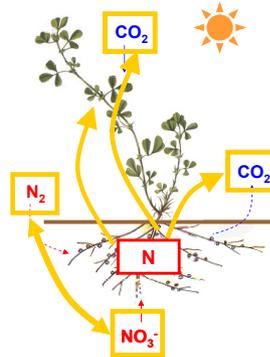
Combine approaches

Phenotyping

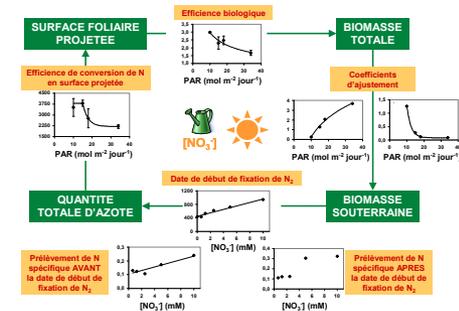


Identify differences
between genotypes

+ Analytical approach

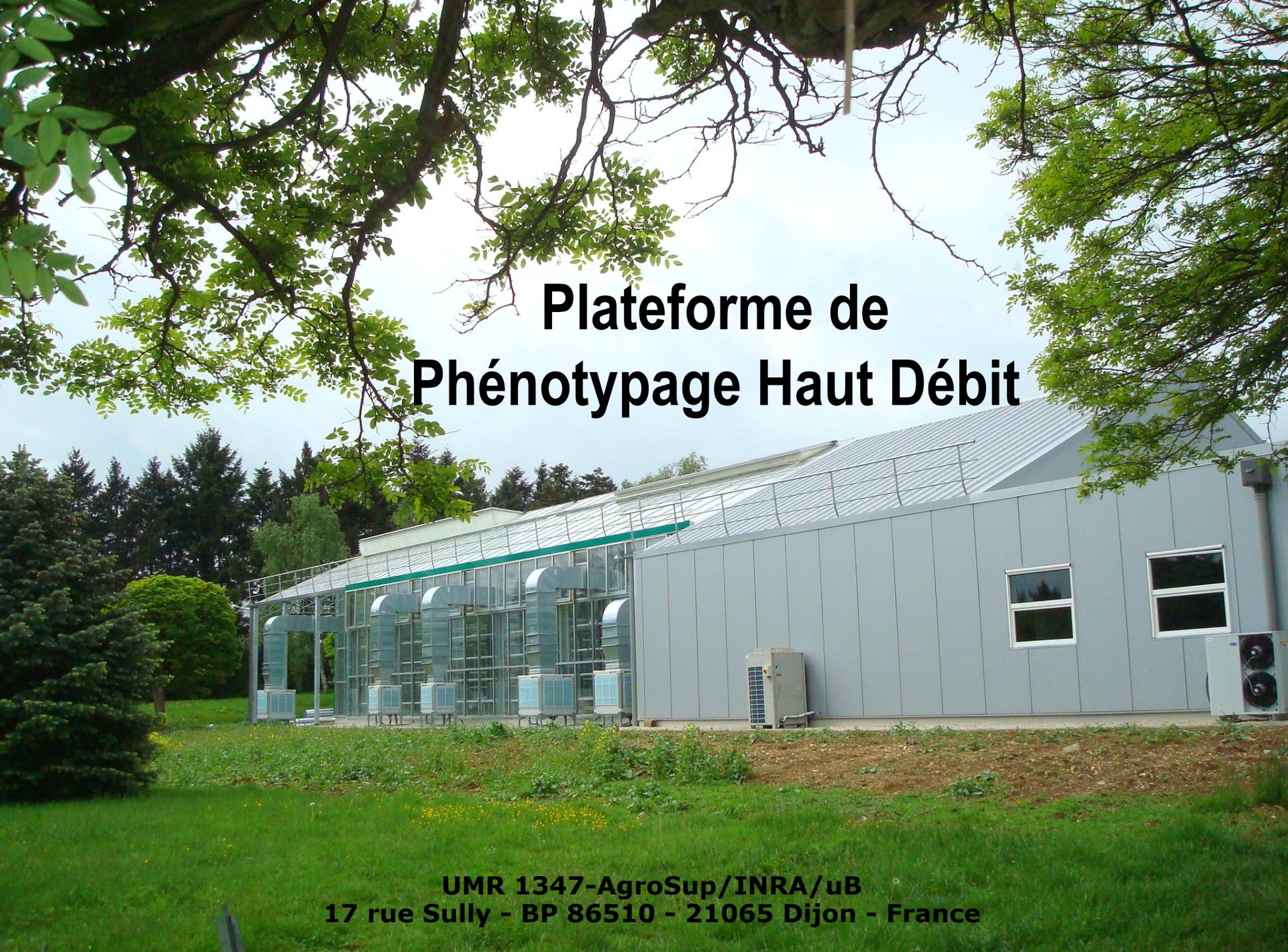


+ Modelization



Interpret detected
differences

Validate in the field to handle G*E*M interactions



Plateforme de Phénotypage Haut Débit

**UMR 1347-AgroSup/INRA/uB
17 rue Sully - BP 86510 - 21065 Dijon - France**

Large Research Unit « Agroecology »

Goals

⇒ A variety of research objects

EcoDur



**Legumes,
Weeds,
Cereals,
Brassica**

GEAPSI



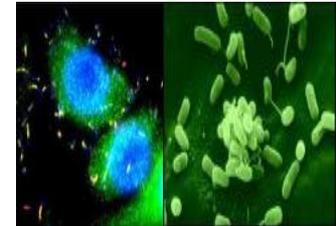
**Legumes,
Legumes/
Cereals
Associations**

IPM



**Legumes,
Arabidopsis,
Wine,
Tobacco,
Tomato**

MERS



**Listeria and
root tissues**

- Produce plant material in characterized environmental conditions.
- Use and develop innovative techniques for high throughput morphometry

... of a large number of biological units,
their interactions...



... under various environmental
conditions...



[CO₂] ↑

... at various organisational levels
(organ /plant)



... and phenotyping equipment

Aerial architecture



20 units/h



Organs (seeds...)



Small plants



6 units/h

Germination



2 units/h



Root system



Licence  INRA 

120 units/h



100 units/h



VIS
NIR
FLUO



Analyze genetic variability by large genotypes series,
various environmental scenarios.

Example: focus on legume plants

Phenotyping plant architecture, flowering, senescence, ...



P. sativum *M. truncatula*

Phenotyping pods and seeds



✓ Seeds : Number, size and shape

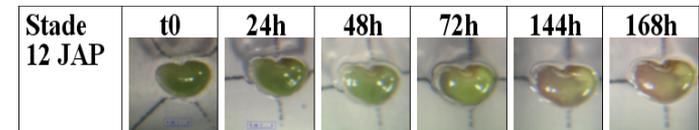


Kroj et al. Development (2003) 130, 6065-6073

✓ Fluorescence: GFP



✓ *In vitro* kinetic development



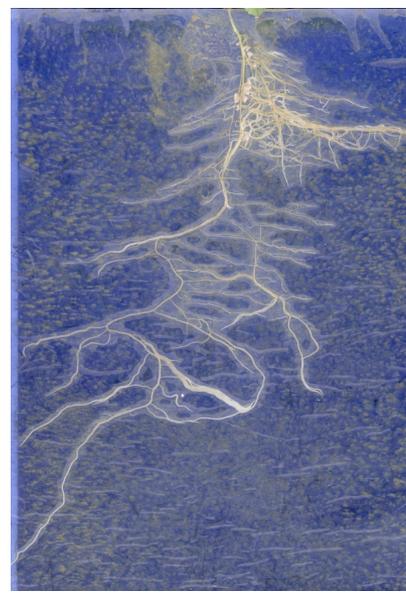
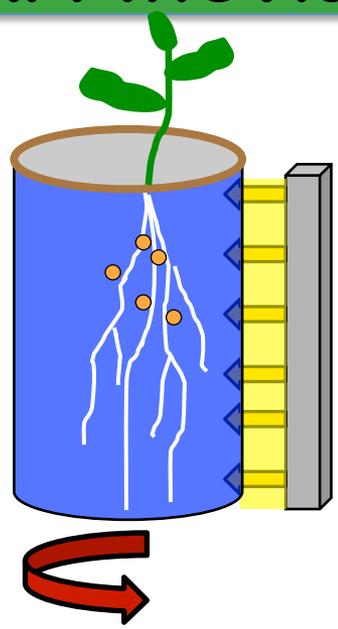
- Mutants (Tilling..)
- RILs
- Ecotypes collections

CONTROLLED ENVIRONMENTAL CONDITIONS



Phenotyping roots and interactions with micro organisms (nodules, μR), ...

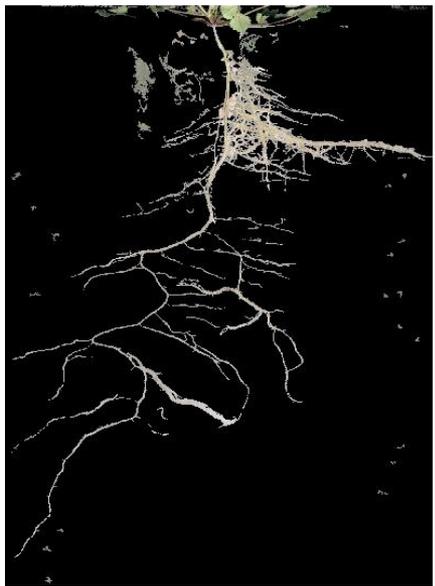
Rhizotron (INRA-Inoviaflow EU licence)



HD Rhizotrons (1300 units planned)
....

Rotating scan

RBG Image



Phenotyping Traits :

- projected root area
- projected nodule area
- total nodule numbers
- nodule size classification
- total root length
-

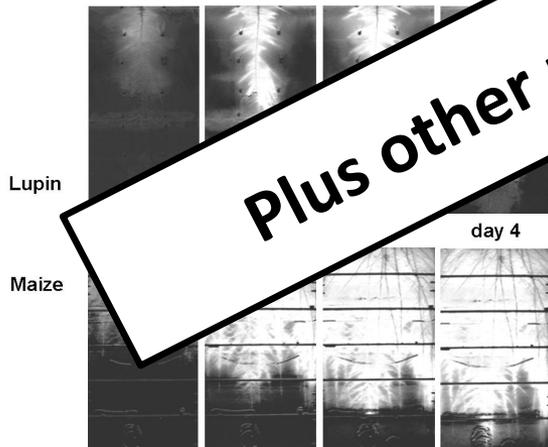


ID	Class	Accession	W. Root	Root	Yield/Plant	Yield/L	Yield	Masses 1	Masses 2	Masses 3	Masses 4
4882	38	001	AB02	Str	Feb	20	2	1252	0.8	79	125
4882	38	001	AB02	Str	Feb	20	4	1260	0.8	79	125
4882	38	001	AB02	Str	Feb	20	6	1244	0.8	79	125
4882	38	001	AB02	Str	Feb	40	80	1254	0.8	79	125
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4882	38	001	AB02	Str	Feb	40	80	1251	0.8	79	125
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4882	38	001	AB02	Str	Feb	40	80	1253	0.8	79	125
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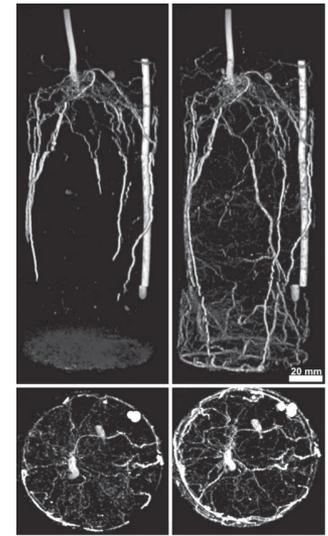
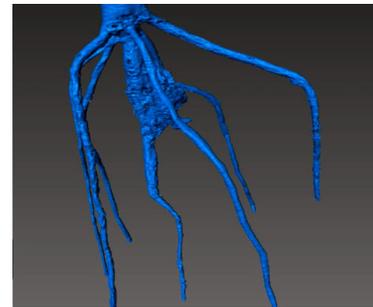
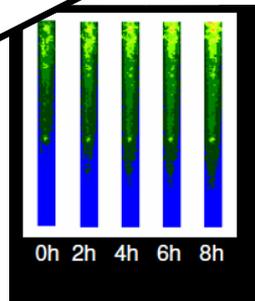
Brief summary of root phenotyping tools

	RGB/NIR	X rays	MRI	Thz
« analysis » window	+++	++	+	++
Analysis length	1 mn	1 mn	20 mm	1 m
Resolution	1 mm	0,2 mm	1 mm	1 cm
Dangers	None	Ionisation	Magnetic field	None
Price	+++	+	--	--
Real time	+++	+	+++	+++

Plus other project for automatization on going



Dry Wet 50 cm **Figure 5**



Phenotyping structuration: local, national

Plateforme Phénotypage Haut Débit



EFOR

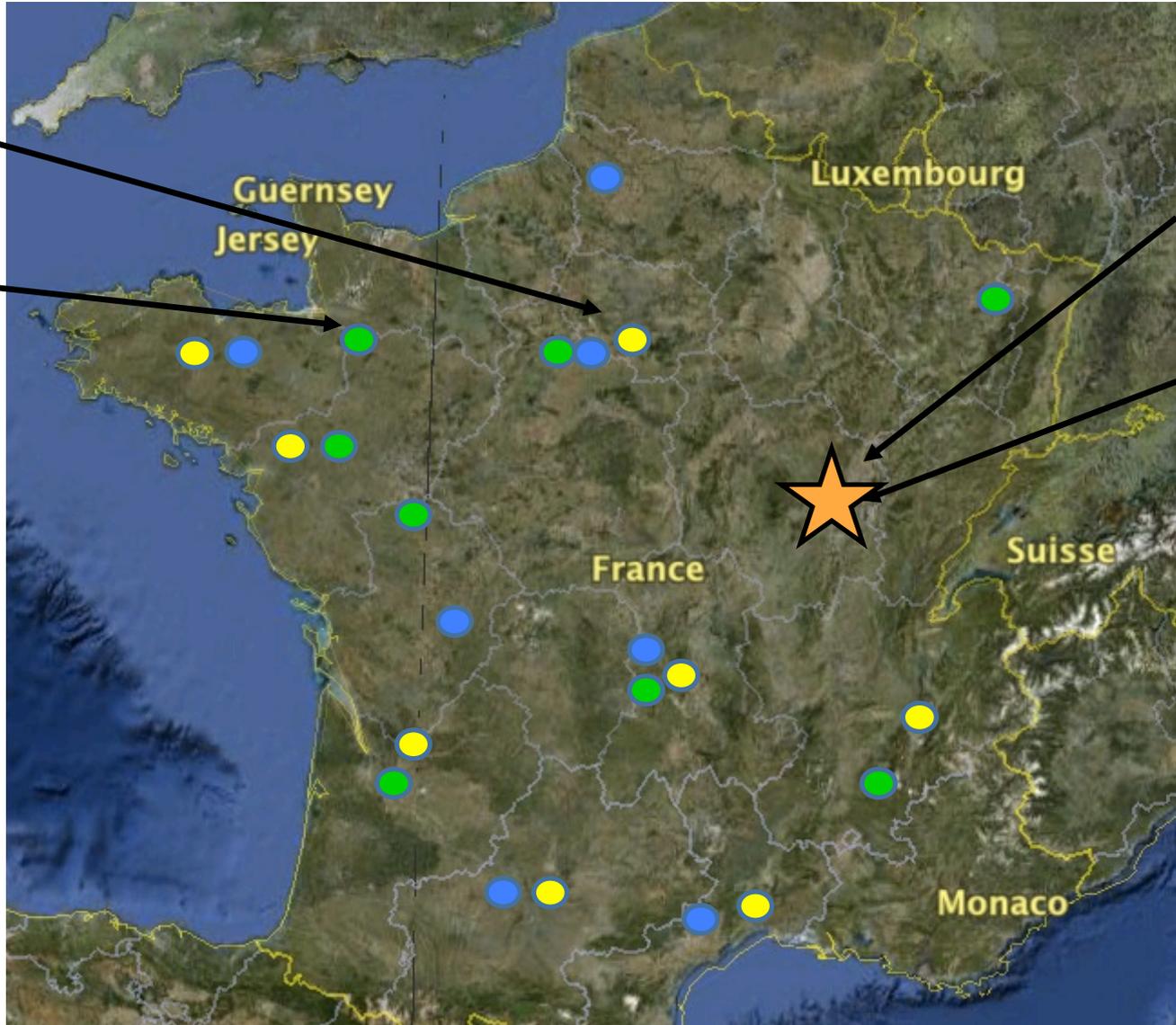
FUI
SERAPIS



CAQ40
ACCAF

PEAMUST

PHENOME



International



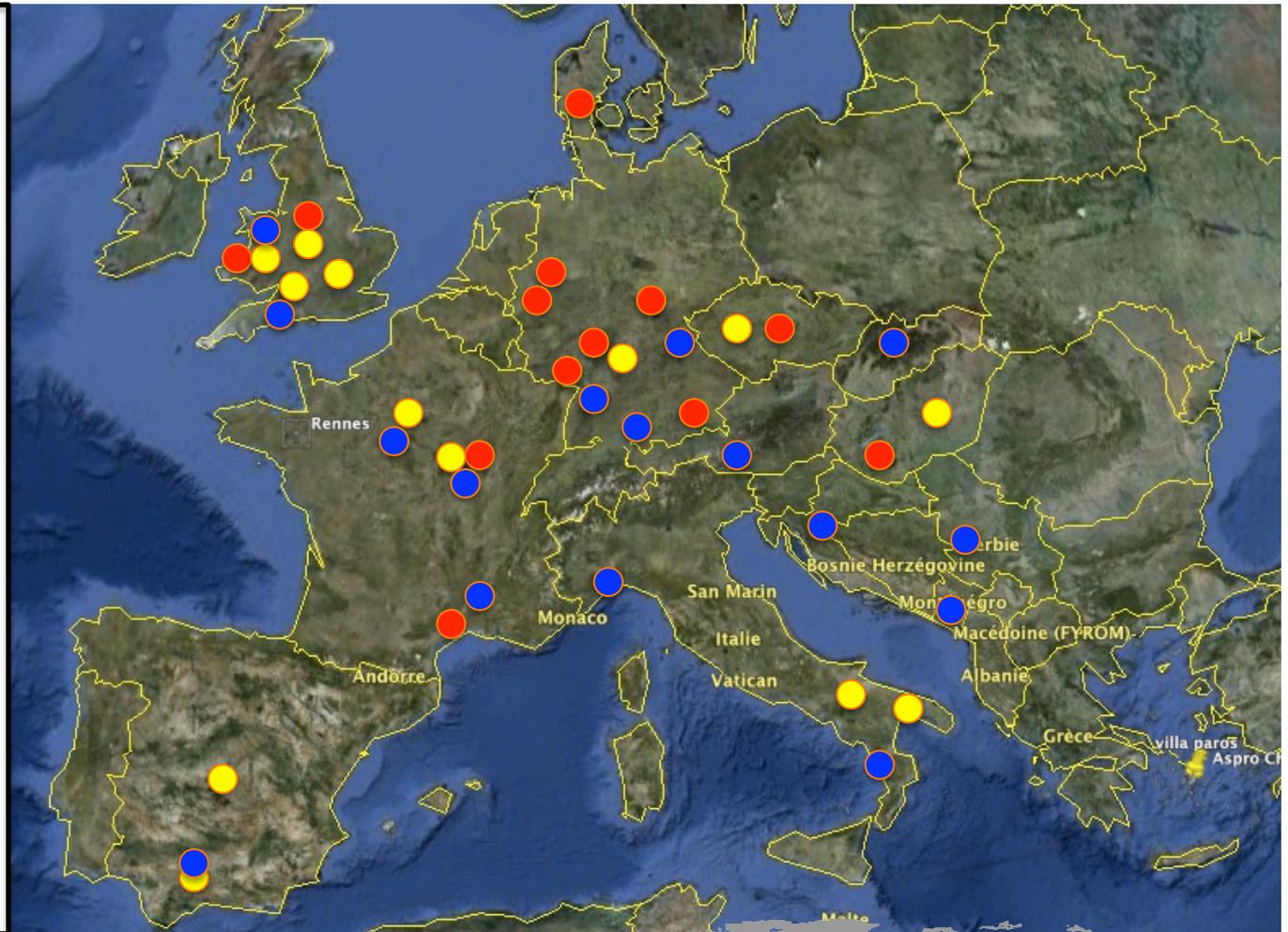
European Plant Phenotyping Network
EPPN

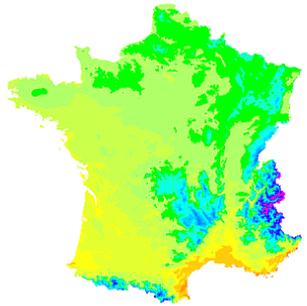


ABS RESS
improving the resistance of legume crops to combined Abiotic and Biotic STRESS



LEGATO



Examples of :

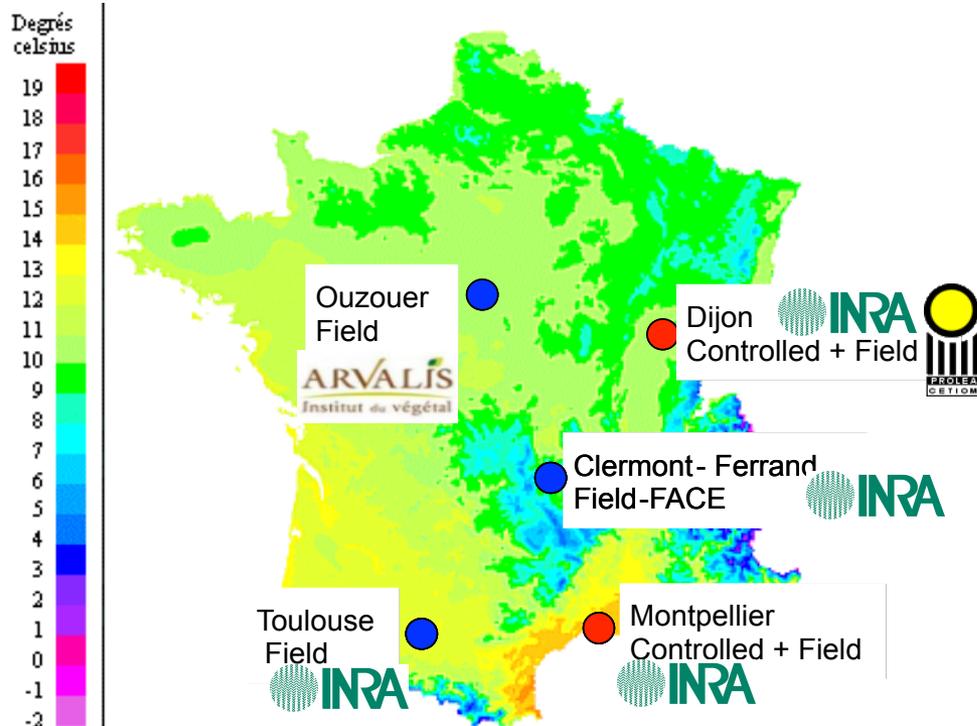
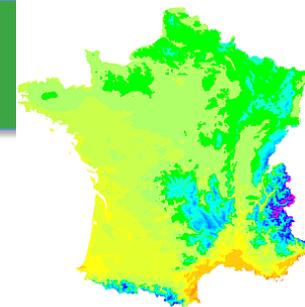
- Phenome: French initiative (30M€)



- EPPN: EU project(5,5 M€)

Prérequisites

- Homogenous high throughput : necessitates 200 – 500 genotypes
“...make on these genotypes what we made on 10 plants...”
- Control / measure environmental conditions encountered by plants
- Measure traits of interest:
 - « ... In platforms (either controlled or field) we'll measure only what we cannot do elsewhere ... »
 - reproducible / heritable / characters involved in yield (its quality), “hidden” (RUE rather than biomass)
 - “genotypic parameters of ecophysiological models”
- Flexibility in order to allow working on various species



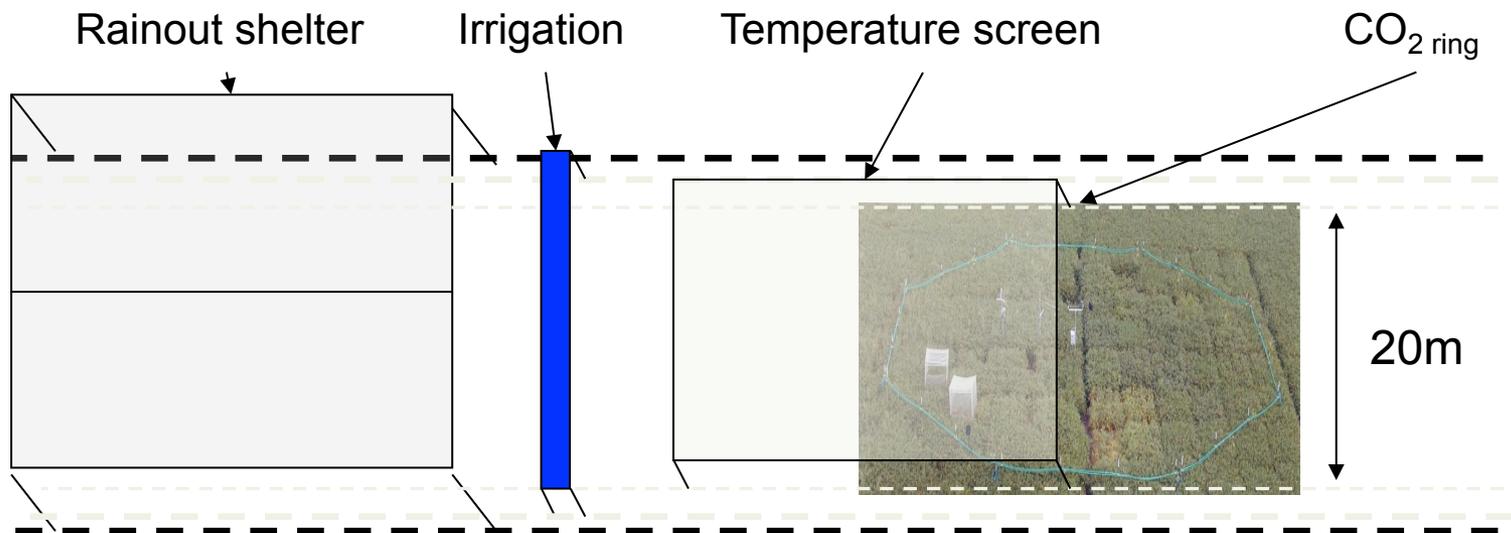
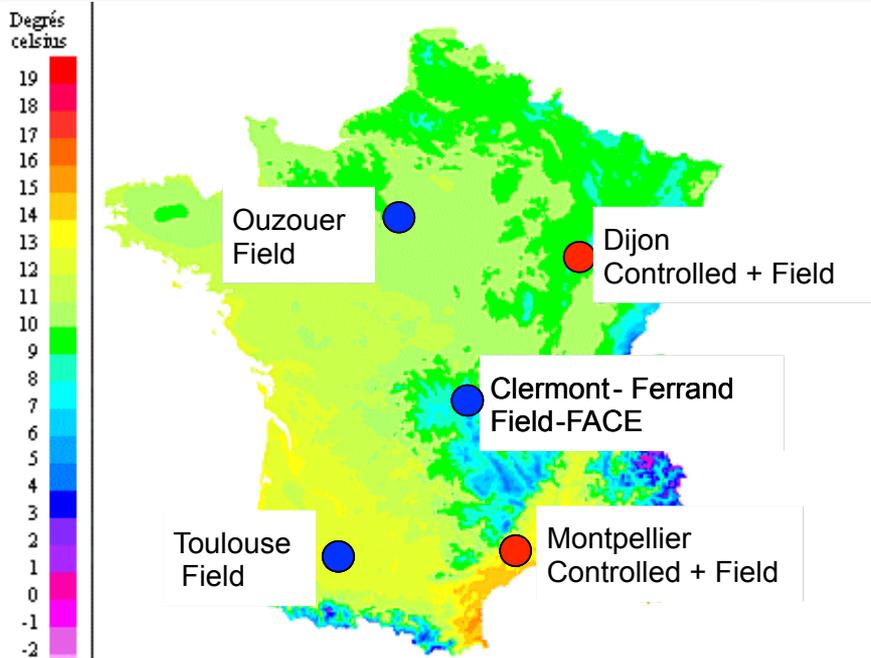
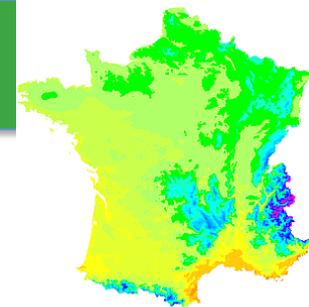
Differents pedo-climatic conditions

Sharing characteristics:

- environnement sensors (soil and air)
- sensors of plant responses
- local database

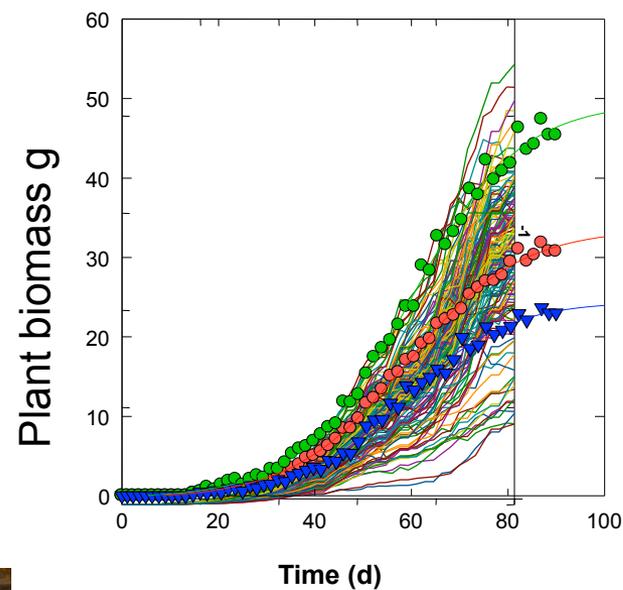
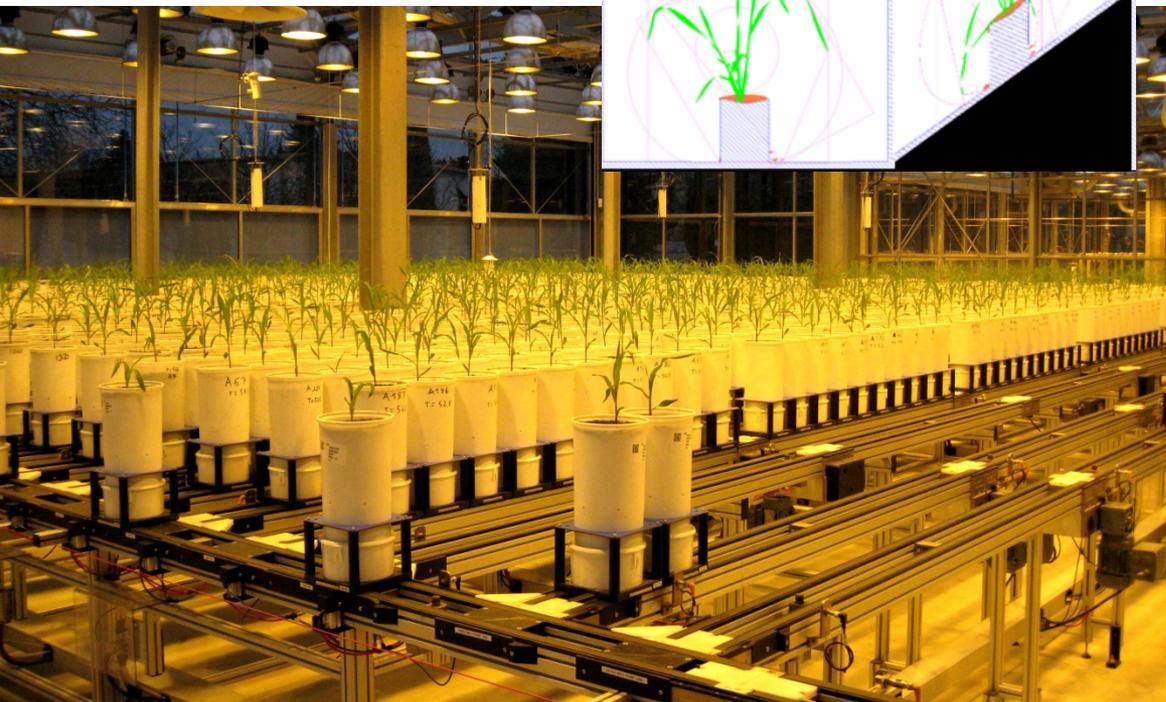
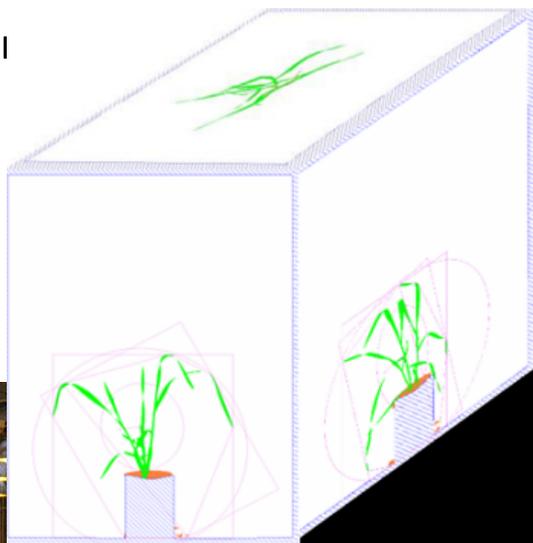
Having specificities :

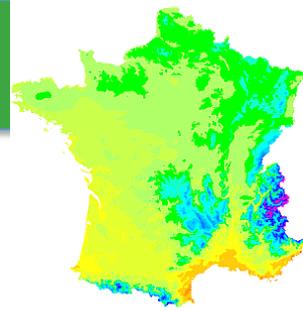
- rolling shelters = water
- FACE = [CO₂]



PhenoArch (Montpellier)

PPHD (Dijon)





Develop new sensor and methods

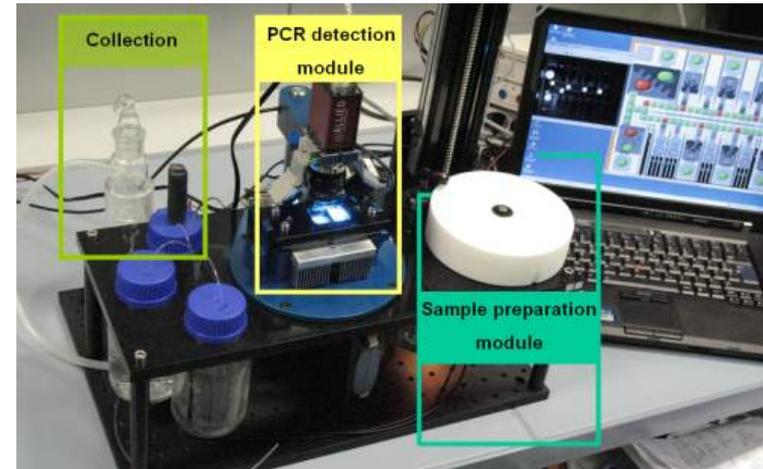
Consortium INRA + INRIA + polytechnique + CEA LETI

Autonomous and miniaturized sensors:

Organ temperature, light



Pathogens presson



EPPN is the first integrated FP 7 EU

Research Infrastructure project in Plant Sciences

Goals:

Create a European integrated network

Provide access to EPPN facilities for the user community

Develop novel instrumentation for non-invasive methods

Establish definition of standards

Duration: January 2012 – December 2015

Budget: 5 500 000 €



Organisation	
Forschungszentrum Jülich, Germany (coordination)	FZJ
Leibniz-Institut für Pflanzengenetik und Kulturpflanzenforschung, Germany	IPK
Helmholtz Zentrum München, Germany	HMGU
Rheinisch-Westfaelische Technische Hochschule Aachen, Germany	RWTH
Institut National de la Recherche Agronomique, France	INRA
Aarhus University, Denmark	AA
University of Nottingham, United Kingdom	UNOTT
Wageningen University and Research, The Netherlands	DLO
Hungarian Academy of Sciences, Hungary	HAS
Austrian Plant Phenotyping Facility, Australia	APPF
Global Change Research Centre, Czech Republic	CzechGlobe
Aberyswyth University, United Kingdom	ABER
Keygene Inc., The Netherlands	Keygene
Phenom-Networks, Israel	Phenom



Thanks for your attention ...