

Phenotyping is the major limitation for selecting genotypes with maintained performances under climate change and reduced inputs.



The Phenome project funded by the "Investments for the Future" program (ANR-11-INBS-0012) and coordinated by F. Tardieu (INRA LEPSE) develops a versatile, high-throughput infrastructure and methods allowing characterisation of hundreds of genotypes of different species under various environmental scenarios. BreedWheat will take advantage of several Phenome platforms.

The Phenome infrastructure consists of **(1) two platforms in controlled conditions** with management of soil water content, temperature and CO<sub>2</sub>; **(2) two field platforms with controlled environmental conditions** including large rainout-shelters and one free-air carbon enrichment (FACE) system; **(3) three field platforms with higher throughput** equipped with soil and climate sensors. All platforms are equipped with a set of 3D functional imaging techniques, namely detailed imaging of roots and shoots in controlled conditions, canopy imaging with an autonomous 'phenomobile' that captures functional and 3D images of each plot, and drones that image hundreds of plots jointly in visible, infrared and thermal infrared bands. **Two supporting "omic" platforms** centralise metabolomic and structural measurements associated with experiments in phenotyping platforms.

**Methodological projects at the infrastructure level develop methods and techniques** able to (1) improve our capacity to measure plant traits at different resolutions in field and platforms (e.g. root and shoot architectures, light interception, transpiration rate) and environmental conditions (novel sensors); (2) organise phenotypic data originating from different platforms, so that they can be saved and analysed for a long period by a wide scientific community; (3) handle very large datasets thanks to data cleaning via artificial intelligence, analyses of time-related data, and interface with plant and crop models

These methods and techniques are to be widely transferred towards the academic and industrial phenotyping community. Phenome resulted in the development of SMEs aimed at phenotyping and is fully integrated to European and international projects (EPPN, IPPN, European roadmap).

For more information on Phenome, please visit :

[www.phenome-fppn.fr](http://www.phenome-fppn.fr)

## More information

[www.breedwheat.fr](http://www.breedwheat.fr)

**Coordinator:** Dr. Jacques Le Gouis, UMR GDEC - jacques.legouis@clermont.inra.fr

**Project manager:** Emmanuelle Lagendijk, INRA Transfert - emmanuelle.lagendijk@paris.inra.fr

**Communication manager:** Grégoire-Yves Berthe, Céréales Vallée  
gregoire.berthe@cereales-vallee.org

This project receives funding from the French Government managed by the Research National Agency (ANR) in the framework of the Investments for the Future (ANR-10-BTBR-03), France Agrimer and the French Fund to support Plant Breeding (FSOV).



## Breeding for bread wheat adaptation to abiotic and biotic stresses

June 2015

**BreedWheat aims at strengthening the competitiveness of the French wheat breeding sector as well as to address the societal demand for sustainability, quality, and safety in agricultural production. In this frame, Workpackage 2 (WP2) combines several approaches as ecophysiological studies, crop modelling, genes network modelling and association genetics to decipher the adaptive response of wheat to major biotic and abiotic constraints.**

### Ecophysiological and -omic studies to identify candidate traits and genes

Ecophysiological and -omic studies have been and will be conducted for a better understanding of plant responses to abiotic (high temperature, limited nitrogen (N) and sulfur (S) or drought) and biotic stresses (*Septoria* and *Fusarium*). Our goal is to identify traits and candidate genes involved in the tolerance to these main constraints.



Nitrogen field experiment

Results will be used to :

- (1) provide a list of candidate genes.
- (2) improve a structure-function crop model that will permit to identify potential plant models called ideotypes
- (3) identify new phenotyping traits/tools

These genes will be identified through differential expression analyses and regulatory gene network mining. High-throughput molecular markers will be developed in WP1 ([see Booklet #2](#)) which will be used in subsequent association studies to identify SNPs linked to traits of interest. **grain development.**

To study high temperature tolerance, grains sampled from plants of two contrasted genotypes submitted to post-anthesis heat stress (29°C) were collected and analyzed. **From anthesis to maturity, grain dimensions, volumes and masses were measured**, as well as water variables (water content, water potential, osmotic pressure). The data indicated a **significant effect of high temperature on grain development.**

On these samples, RNA were extracted from 11 grain developmental stages. The analysis of the data corresponding to 88 hybridizations identified several sets of genes whose expression is modulated by temperature. For example, 1733 genes are impacted by high temperature in a "resistant" genotype while 2606 genes are impacted by temperature in a "sensitive" genotype. These genes will be candidates for an *a priori* approach in association studies.

Concerning limited N and S stress, a main achievement was the production of samples to understand molecular mechanisms in relation to N use efficiency (NUE) for grain yield and protein concentration. Preliminary results based on the composition of mature seeds confirm the impact of these treatments. Samples from seed at different developmental stages were harvested for further -omics analyses which will identify candidate genes for NUE and will be used in an *a priori* approaches in association studies.

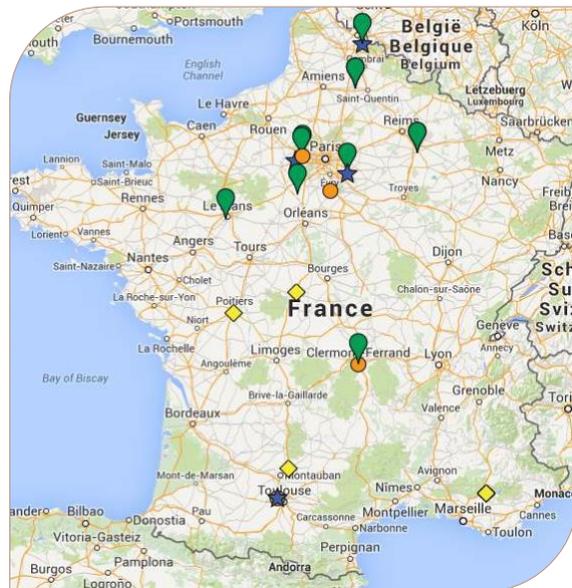
In addition, RulNet, a tool to infer and analyze regulatory networks from qualitative and quantitative -omics data was developed as a web-oriented platform ([wheat-urgi.versailles.inra.fr/Tools/RulNet](http://wheat-urgi.versailles.inra.fr/Tools/RulNet)).

Finally, two experiments were performed in glasshouse to investigate *Septoria* development in canopies with identical structure and contrasting leaf N content. Results showed a strong effect of leaf age and leaf senescence on *Septoria* development.

## A network of field experiments for quantitative studies

Quantitative studies have been and will be conducted to identify genomic regions involved in adaptation to abiotic and biotic stresses. A network of 25 field trials was set up over 3 years and focused on three major constraints : limited N, drought and diseases (*Septoria* and *Fusarium*).

A panel of 220 winter European elite varieties (BWP2 panel) was tested on each trial in normal and stressed conditions. This genetic material was genotyped using a 420K SNP Axiom array (TaBW420k) developed within BreedWheat WP1 (see Booklet #2) leading to 92K polymorphic SNPs. Several traits were scored on these field trials, from grain yield and grain protein concentration to yield components and biomass.



BreedWheat trials mapping

New phenotypic traits like NDVI (Normalized Difference Vegetation Index) were also tested. Environmental characterization of each field trial was performed to validate the presence of a significant stress, particularly for drought trials.

Agro-climatic data were then analyzed to determine the period and intensity of each stress. A working group « Data Integration and Analysis » was created to define a strategy for the analysis from the raw data to association results. A workflow of analysis was then built to produce whole genome association results for each trial. Genotype x Environment interactions will be also analyzed.

Finally, a meta-analysis will be performed to select 20 regions of interest for fine mapping and 5 of them for field validation.

## Database, new panel and new phenotyping methods

WP2 has produced a large quantity of results, data, and samples during 3 years.

Exchanges with the other BreedWheat WPs are very important: with WP1 for genotypic data, with WP5 for implementation in the database BreedWheat Information System (BWIS) of phenotypic data and association results. Interaction with WP3 led to the selection of lines for the new diversity panel BWP3 (see Booklet #1) which will be phenotyped in 2016/17 and used to perform association and cross-validation of our association results obtained on BWP2 panel (elite lines).

Before that, the BWP2 panel will be phenotyped again in 2015/2016 in two field controlled conditions for drought stress using rainout shelters to manage water availability (Arvalis and INRA platforms built within the Phenome project (see below)). These high throughput phenotyping platforms will give us access to new types of phenotypic data using sensors along the entire plant development cycle.



Phenofield platform

For the coming years, valorization of all these data will be done and we hope to identify relevant genetic/genomic areas linked to traits of interest. Our goal is to develop informative markers to facilitate selection for stress tolerance and also to select the most promising regions for fine mapping and to validate their effect in other materials in field experiments.

For more information on BreedWheat WP2, please contact :

Stéphane Lafarge  
from Biogemma  
[stephane.lafarge@biogemma.com](mailto:stephane.lafarge@biogemma.com)