Transnational Access Report

1. General Information

Project Acronym (ID):	Dwarfdrought
Project Title	Destructive and non-destructive quantification of drought resistance in isogenic tall and dwarf wheat
Installation used	4PMI
Name of Group Leader	Josh Klein
Name of organization	ARO-Volcani Center Bet Dagan, Israel

2. Project summary (max. 250 words)

Dwarfing genes in wheat not only reduce lodging and increase grain yield but also enhance plant resistance to water stress. Investigating the physical attributes and the physiological mechanisms associated with drought resistance is often a time-consuming and destructive procedure, especially when studying roots. We investigated automated phenotyping of roots and shoots as a non-destructive method for quantifying physical and physiological aspects of drought resistance intall (rht) and dwarf (Rht3; *Rht-B1c)* Berséewheat seedlings. During a month of growth, plants were exposed to no, medium, or severe water stress for 10 consecutive days, corresponding to 100, 60, or 40 % of field capacity at the end of the stress period, followed by a return to full irrigation for a week of recovery. Plants were made available on conveyors equipped greenhouse for phenotyping throughout the experiment: this involved at some time points destructive and non invasive measurements and analyses and non destructively. A series of three experiments was performed to setup the protocol, train on methods, adjust experimental conditions then a larger experiment was finally developed. All experiments were carried out at the highly-automated Plant Phenotyping Platform facility at INRA, Dijon.

3. Main achievements (max. 250 words)

The best model for calibration with destructive measurements of plant compartments area and biomass was obtained by taking three images of projected shoot area. Using the curve parameters linking the projected area with measured (i.e. destructive) shoot area allowed us to dynamically calculate non invasively shoot fresh and dry weight for further dynamic, non destructive characterization. Plant heigh simulation was more tedious due to overlaping and bending leaves. The proportional effects of moderate or severe water stress in decreasing plant height and leaf area were comparable, whether measured destructively or not. Growth could therefore be measured continuously, rather than at static intervals. Even after a week of recovery, overall leaf area was reduced 25% in moderately-stressed and 60% in severely-stressed plants, although dwarf plants fared better than tall. Plant height recovered more rapidly than leaf area, with no residual effect found in moderately-stressed plants, and a 30% reduction in severely-stressed plants. The proportional effect of stress on both leaf area and height could be quantified automatically using three lateral views of shoots, which limited under-estimation resulting from leaves masking each other. Leaves that exceeded 25 cm often bent over,while some never grew taller than 5 cm. Despite calibration, in these extreme cases leaves could be miss-measured by as much as two-fold. Technical fixes for these problems are being developed in both hardware and software.

The number of main roots per seedling decreased 15-25% under moderate or severe stress, while the number of secondary roots increased 33% under severe stress. Main roots of dwarf wheat were 30%

longer under moderate or severe stress, but secondary roots were up to 37% shorter, especially at the growing end of the root. Images from the rhizotron were analyzed manually using the programs ImageJ or Dart, since wheat roots were often too translucent and too intertwined for quantification by automated machine vision. Chlorophyll, carotenoid, and anthocyanin concentrations in leaves changed with degree of water stress, and also in response to seed treatment with trinexapac-ethyl, which induces resistance to stress. We were unable to correlate leaf pigment values with R, G, B and hue values extracted from RGB color images.

Leaf area is a good indicator of water stress in wheat.

Dwarf (Rht) or dwarfed (TE) wheat recovers better from water stress than do Tall (null) or control plants

Roots of dwarf plants resist water stress dynamically – changes in main root length and secondary root formation: last experiment is on-going

Leaf color does not express pigment concentration.

The automated methods that have been developed allowed efficient acquisition and analysis of data from individual wheat plants in 250 pots (shoots) and 120 rhizotubes (roots). Continued improvement of these methods will greatly increase possibilities and precision in large-scale experiments with agronomic and horticultural crops.

8. Publications related to the access granted, acknowledging the support by EC.

Destructive and non-destructive quantification of drought resistance in isogenic tall and dwarf wheat

Joshua D. Klein, Christian Jeudy, Mathieu Chanis,Mickael Lamboeuf,Joshua Herskovitz, YonitRazShalev, Christophe Salon. EPPN general meeting, Barcelona, 10-11th of November, 2015.