

Dear all

Enclosed please find an announcement of a new Phd-project (both the formal announcement and a small resume). Its a joint project between AU and KU/LIFE so the supervisors are Sven Bode (prof. In genetics), Eva Rosenqvist and I.

If you have any suggestions for a potential candidate with knowledge about photosynthesis and genetics - please spread the information or announce it through your announcement system.

Best wishes

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Phd-project

## ***Wheat in (dis)stress***

### **Objective**

The objective of the Ph.D project is to identify the physiological and genetical mechanisms for wheat to maintain a high and stable production under conditions of variable climate. Key traits being considered are water-use efficiency, photosynthetic light- and carbon-use.

### **Hypothesis**

We hypothesise that the existence of genetic variability in the wheat species can considerably increase tolerance to both heat and drought stress. We further hypothesize that these traits can be detected by phenotyping of photosynthesis parameters and mapped in their unadapted genetic backgrounds (spring wheat) and crossed into existing high-yielding winter wheat lines to obtain tolerance to heat and drought stress and thus resilience to climatic variability.

### **Background**

The major challenge for sustainable crop production globally is climate uncertainty and the increased frequency of extreme climate events (IPCC, 2007). Anticipated changes in climatic variability leading to more frequent extreme conditions will require directed adaptations of crop species on an unprecedented scale in order to sustain agricultural production (IPCC, 2007; Wollenweber et al., 2005). In North Europe crop production will need to cope with more extreme droughts and heat waves (Olesen et al., 2009). In the last decade, severe drought and heat have led to significant yield losses in parts of Europe (Schär et al., 2004, Ciais et al., 2005). These studies demonstrate the need for and potential of more drought and heat tolerant crops in the future.

Wheat is a major crop and accounts for ca. 30% of the global cereal production. Both heat and drought affect grain yield at almost all growth stages, with the flowering phase being the most susceptible stage. The response of plants to water deficit involves both short-term molecular and physiological responses as well as long-term structural and morphological

modifications (Chaves & Liveira, 2004, Wollenweber et al., 2003, Winning et al., 2009, Fast-Seefeldt et al., 2009).

Diverse stress conditions activate both mechanisms of acclimation and adaptation by the plant. Adaptation is related to heritable modifications. Molecular control mechanisms for abiotic stress tolerance are based on the regulation of stress-related genes (e.g. encoding antioxidants and heat-shock proteins). Acclimation is defined as the generation of non-inheritable modifications that reflect the structural and physiological plasticity of the plant to cope with stress.

High temperature and changed water-use will change the source/sink balance of carbohydrates in the plant modifying CO<sub>2</sub> use efficiency. Such dynamic relations are not widely studied but are of significant importance for growth and yield, yet crops often operate far from maximum capacity (Murchie et al., 2009). The gas exchange capacity and the stomatal regulation are expected to vary in response to stress.

More information is needed on the molecular mechanisms of physiological responses of wheat to water deficits as well as on the consequences for wheat yield and quality (Wollenweber et al., 2005). Advances include improved genetics with enhanced marker technology and QTL detection methods, complex trait physiology with prediction of consequences of genetic variation, and advances in modelling linked to QTLs.

Conventional breeding of wheat has relied upon repeated recombination of adapted material to search for relatively small improvements. Introduction of new genes from unadapted material to the high yielding gene pool has mostly been inefficient, particularly for complex traits like adaptation to stress tolerance, which cannot be selected efficiently by conventional means. However, there is recent evidence that increased stress tolerance and yield can be introduced from wild relatives through genetic mapping and marker assisted backcrossing (Mallikarjuna et al., 2008). At the same time such genetic differences affecting complex traits like drought and heat tolerance can be useful for elucidating the underlying molecular mechanisms. Studies of cosegregation of genes with advanced physiological measurements or metabolomics is a widely applicable approach (Schauer, 2006) and cloning of such genes for direct studies of transcript and product function is possible in wheat (Fu et al., 2009, Krattinger et al., 2009). Conventional backcrossing for introgression of the genes is generally hampered by inefficient initial selection for adaptation in early generations, so that undesirable linked genes are cotransferred. For these reasons advanced physiological measurements of stress responses to substitute for yield measurements in the genetic mapping process are likely to provide new dimensions to this technology (Yin et

al., 2009) and the effect of such physiological traits can be evaluated using state-of-the-art mathematical crop models (Sinclair and Muchow, 2001, Campos et al., 2004).

Northern Europe currently has a cool and moist climate conferring high yield potential. Consequently cultivars have been developed that are suited to these conditions. However, these cultivars have a low tolerance to heat and drought. Tolerant varieties may be found in cultivars adapted to dry and hot regions. Synthetic wheat lines include combinations of tetraploid *Triticum durum* and the wild *Triticum tauschii*, both mostly adapted to South Europe. Both species are supposed to have a high degree of drought and heat tolerance. These assumptions need to be tested both through genetic and physiological screening.

## **Methods**

Chlorophyll *a* fluorescence is a fast, non-destructive method for screening stress tolerance (Baker and Rosenqvist, 2004), when combined with measurement of gas exchange the degree of stress can be quantified. The project will only use existing and naturally occurring genes.

1000 synthetic wheat lines derived from crosses between *T. durum* and *T. tauschii* will be established from collections in CIMMYT and ICARDA. An additional 900 cultivars of spring wheat from Europe and the near East are currently under multiplication at KU-LIFE. Both sets of material will be available for the first physiological screening to identify lines with superior tolerance to heat and drought.

Promising synthetic lines from the physiological screening (WP1.a) will be backcrossed with high yielding lines to BC1. The segregating single plants of the BC1 will be evaluated and mapped with a set of SSR markers spanning the entire wheat genome followed by identification of the genes with QTL analysis. Further backcrossing into several high-adapted wheat lines and marketing will be performed in collaboration with breeding companies.

Large populations of offspring (ca. 1500-2000 plants) will be generated as BC1 from combinations segregating for interesting genes for stress tolerance. These populations will be used for detailed physiological investigation through co-segregation of physiological measures with the linked markers. For particularly interesting genes 200-250 recombinants will be selected with flanking markers, evaluated with physiological measurements and close mapped. To saturate the chromosomal region surrounding the QTL we will select 100-150 genes from the synthetic segments of rice and *Brachypodium* and use the wheat EST database to design PCR primers spanning introns (Ishikawa et al., 2007; 2009). After PCR of

the intron areas, the PCR products will be bar coded for each plant separately and sequenced with 454 sequencing (outsourced). The sequences will be searched for SNP (Frei et al., 2009) between parents linked to the QTL to find closely linked markers for improved marker assisted selection and co-segregation studies for physiology. Important genes may subsequently be map based cloned.

The effect of high temperature at a fixed light level will be measured as the maximum quantum yield of PSII,  $F_v/F_m$ , which is a fast (seconds) screening method. The susceptibility to drought can be detected by the effective quantum yield of PSII,  $\Delta F/F'_m$ . Fluorescence quenching analysis during stress will be applied to obtain mechanistic information of the energy balance of the leaf in terms of photochemistry and protective heat dissipation (Rosenqvist, 2001).

### **Time schedule**

The entire project in collaboration with project groups of plant physiology and molecular genetics is scheduled for three years.

### **Phd-courses**

The phd study in addition to the project work will include approx 6 months of phd course activity.