APPROVED SUBCONTRACTOR PROJECT:

Exploring the use of $\Delta^{18}O$ and total mineral ash content in wheat as a new tools for phenotyping wheat with respect to water inputs – John Foulkes, University of Nottingham

Background:

Drought is an environmental factor that constrains the yield of all crops. In wheat drought conditions can impact yield throughout the life cycle of the plant, although lack of water during grain filling has a particularly severe effect. Breeding for drought resistance ultimately relies on the ability to select for yield under reduced water input; the measurement of secondary traits, such as senescence (stay green) and high vegetative biomass that are correlated with sustained grain yield under such growing conditions can be used to identify germplasm with potential for drought tolerance. Carbon isotope discrimination ($\Delta^{13}C$) has been used in wheat to screen for genotypes with high transpiration efficiency[1] and this has led to the release of two wheat varieties ("Rees" and "Drysdale") in Australia that are better suited to drier conditions during grain filling. More recently another isotope-based screen using ^{18}O enrichment has been shown in maize[2] and wheat[3] to correlate with yield potential and drought resistance[4]. The $\Delta^{18}O$ signature of vegetative tissues is known to reflect variation in evaporative enrichment in leaves due to transpiration[5], and has been shown to be negatively correlated with transpiration rate (Barbour et al., 2000). Therefore, measurement of the $\Delta^{18}O$ signature might provide a powerful tool for plant breeders to track genotypic differences in drought resistance. In addition, the total leaf ash content of plant tissues has been suggested as a useful tool to predict yield performance under drought[6]. The mechanism of mineral accumulation in plant tissues appears to be explained through the passive transport of minerals via xylem driven by transpiration. Thus, ash content measured in plant tissues provides an indicator of transpirative gas-exchange activity and therefore of the total water transpired. The analysis of mineral ash content analysis is also significantly less expensive than $\Delta^{13}C$ or $\Delta^{18}O$, so may be a useful alternative for these screens in breeders' trials.

In this study we will explore the use of $\Delta^{18}0$ and total mineral ash content in wheat as new tools for phenotyping wheat with respect to water inputs. We will study a panel of wheat genotypes for which there are existing data on drought tolerance (at Nottingham and from the SA LINK project based at Broom's Barn). We will compare genetic variability in $\Delta^{18}0$ (leaf) and total mineral ash content (leaf and grain) with that for $\Delta\ ^{13}C$ (leaf and grain) and water use (from direct measurements of volumetric soil water content using capacitance probes in the trials) in the WGIN drought trials at Sutton Bonington in 2009/10 and 2010/11.

In summary, the $\Delta^{18}0$ value is expected to provide added value to the WGIN drought trials (in addition to the existing delta $\Delta\ ^{13}C$ value being measured) because, although much attention has focused on improving WUE when breeding for drought adaptation, it seems that, except for very severe drought conditions, water use (WU, i.e. the total water absorbed and further transpired by the plant) is a more important adaptive trait than WUE[7]. This is related to the genotypic capacity to use available water and therefore to sustain transpiration under unfavourable conditions. The $\Delta^{18}0$ of vegetative tissues can be used as an indirect measure of transpiration and WU.

In addition, ash content of plant dry matter potentially offers a technique to predict WU and yield and genotypic adaptation to drought in an inexpensive way. This method has not been explored to date in our trials since we have favoured the more established Δ C13 technique as indicator of WUE. However, with a view to phenotyping much larger association genetics panels in future projects where costs may become prohibitive, it is worth testing relationships between ash content and genotypic differences in WU and grain yield in the present WGIN2 drought trials. If results are encouraging, this technique could then be applied more widely in future prebreeding research activities as well as in breeders' trials.

[1]Rebetske GJ et al., (2002) Crop Sci., 42, 739-745.
[2]Barbour MM, (2007) Functional Plant Biol., 34, 83-94.
[3]Araus JL et al. (2001) In: Reynolds, M.P., Ortiz-Monasterio, J.I., McNab, A. (Eds.), Application of Physiology in Wheat Breeding. CIMMYT, Mexico, pp. 59-79.
[4]Cabrera-Bosquet et al. (2009) Plant, Cell and Environment doi :10.1111/j.1365-3040.2009.02013.x
[5]Farquhar GD, Lloyd J. (1993) In: Stable isotopes and plant carbon–water relations—Ehleringer JR, Hall AE, Farquhar GD, eds. San Diego: Academic Press, Inc., pp. 47–70.
[6]Araus JL et al. (2002). Ann. Bot. 89, 925–940.
[7]Slafer GA, Araus JL. (2007) In: Spiertz JH, Struik PC, van Laar HH. eds. Scale and complexity in plant systems research: gene–plant–crop relations. Dordrecht: Springer, pp. 145–154.

Workplan:

Plant material and experiment treatments

The plant material used would be that already included in scheduled experiments investigating trait identification for drought tolerance in the activities of Activity 9 of WGIN II (drought tolerance). Specifically, the following the experiments characterising elite cultivars would be used for the investigation of Δ^{18}O and total mineral ash content as drought tolerance screens in each of 2009-10 and 2010-11.

Evaluation of elite varieties

A wide range of genotypes (18), including current and recent elite varieties and parents of DH populations, will be characterized for WUE and drought-tolerance traits under irrigated (trickle irrigation) and unirrigated (rain-fed) conditions in field experiments at University of Nottingham (UN) in each of 2009/10 and 2010/11. In each year, one experiment (2 irrigation treatments x 18 cultivars x 3 replicates; plots size 12 x 1.6 m) will be located on a light sandy loam soil under irrigated (trickle irrigation) and unirrigated (rain-fed) conditions at University of Nottingham.

Proposed measurements of leaf Δ^{18} O and leaf and grain total mineral ash content

In the experiments outlined above, current WGIN II funding covers measurements of grain yield and yield components, grain Δ^{13}C and stem carbohydrate reserves (chemical analysis, anthrone method), and measurements of senescence kinetics in all genotypes as well as water uptake (gravimetric soil analysis) in selected genotypes.

The sub-contract funding would be used to supplement these assessments with the high-throughput screens of (i) leaf Δ^{18} O and (ii) leaf and grain total mineral ash content. In each of 2009-10 and 2010-11 in all plots (108), flag leaves at flowering would be sampled, dried to constant weight and submitted for chemical analysis for Δ^{18} O and total mineral ash content. At harvest grain samples from each plot would be submitted for chemical analysis of mineral ash content. In addition, Δ^{13} C would be assessed on leaf samples at flowering (currently not funded under WGIN II).

Relevance to WGIN objectives

In WGIN II this programme of work will directly relate to the “Targeted Traits” section of “Resource Development”, specifically “improvement of water use efficiency and drought tolerance traits”. We will evaluate the potential of $\Delta^{18}0$ and mineral ash content as new phenotypic screens in wheat for adaptation and yield potential under varying levels of water availability. These studies are also complementary to ongoing studies in the SA LINK project “Improving water use efficiency and drought tolerance in UK winter wheats” (RD-2005-3233; 2006 -2009) and to those in Activity 9 of WGIN II, This proposed screen have not to date been assessed on winter wheat on UK germplasm.