

### Defra Wheat Genetic Improvement Network – Improving the environmental footprint of farming through crop genetics and targeted traits analysis

#### **Background**

The UK government is committed to more sustainable agriculture but this vision is facing an ever expanding range of environmental, energy and climate change challenges. Wheat is grown on a larger area and is more valuable than any other arable crop in the UK. Established in 2003, Wheat Genetic Improvement Network (WGIN) arose directly from a realisation in the early 2000s that over the preceding two decades there had been a widening disconnection between commercial plant breeding activities and publicly funded plant and crop research. The overall aim of WGIN is to generate pre-breeding material carrying novel traits for the UK breeding and deliver companies to accessible technologies, thereby ensuring the means are available to produce new, improved varieties. An integrated scientific 'core' which combines underpinning work on molecular markers, genetic and genomic research, together with novel trait identification, are being pursued to achieve this goal. The programme is managed by a team including representatives of the key UK research groups and breeders. They ensure programme and its outputs are communicated to the wider scientific and end user communities, via a web site, a stakeholder forum, focused meetings and peer reviewed publications. WGIN liaises with equivalent operations overseas to ensure programme is internationally the competitive.

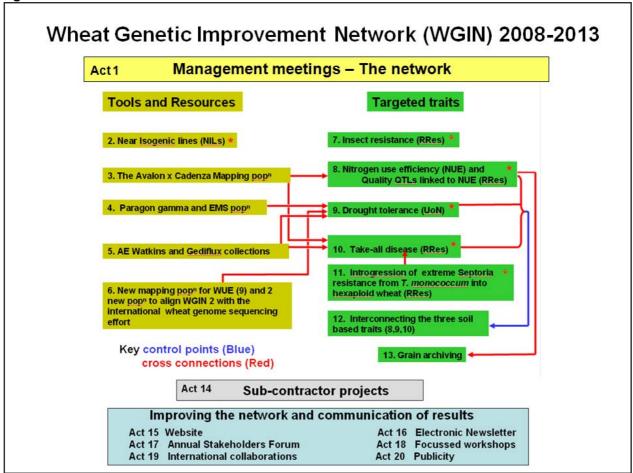
The initial WGIN project ran for five years (2003-2008) and achieved considerable scientific success. In addition, the sustained networking activities and the availability of datasets

generated by the project led to the establishment of many new wheat genetic improvement projects, including some funded jointly by the public sector and industry. Those funded by early 2008 were summarised in the May 2008 Stakeholders Newsletter and since then two additional projects have been agreed. There is no doubt that WGIN has a direct and significant impact on re-establishing the relationship between commercial plant breeding activities and public funded plant and crop research. However significant hurdles remain which currently prevent commercial implementation of much research which should help to reduce the energy requirement and environmental impact of the UK wheat crop.

#### This project

The new WGIN Core Project started in 2008 to provide genetic and molecular resources for research in other defra projects and for a wide range of wheat research projects in the UK. The resources under development include wheat genetic stocks, mapping populations, molecular marker markers and technologies. identification and evaluation, genomics bioinformatics. The twelve "core" research objectives and their interconnections summarised in Figure 1, alongside the six research platform activities designed to promote the further integration of the funded work. initially funded partners (Table 1) are the John Innes Centre, Rothamsted Research and The University of Nottingham but support has been allocated within objective 14 for sub-contracted projects which will be awarded in open competition during 2009 (see website for details). For those familiar with the original WGIN project, a summary of the objectives carried forward from the first WGIN project and those introduced as part of the new project is given in Table 2.

Figure 1: WGIN 2008-2013



Funded partners	Researchers
	Peter Shewry
	Kim Hammond-Kosack
Rothamsted Research	Malcolm Hawkesford
	Richard Gutteridge
	Ruth Gordon-Weeks
	John Snape
	Simon Griffiths
John Innes Centre	Michelle Leverington-Waite
	Simon Orford
	Liz Sayers
	John Foulkes
University of Nottingham	Mattew Tovey
	Research technician (to be appointed)
Subcontractor projects	to be appointed

**Table 2:** Summary of the objectives carried forward from the first WGIN project and those introduced as part of the new project

WGIN 1	WGIN 2	Remarks		
Project management	1. Project management	continued		
Maintaining the network	Maintaining the network	continued		
A. Resource development	A. Resource development			
3. COS Markers		not continued		
	2. Production of near isogenic lines for key traits	new		
3. Mapping populations	3. The Avalon x Cadenza mapping population	continued		
Plant genetic resources and 7 mutagenesis	4. Paragon gamma and EMS mutant lines	continued		
4. Hexaploid diversity screen	5. AE Watkins and Gediflux Germplasm Collections	continued		
	To develop new mapping populations which will align WGIN 2 with the International wheat genome sequencing effort	new		
11. Exploring cereal synteny	QTL development and candidate gene selection for various trait	new		
	B. Targeted traits			
	7. Resistance to cereal aphids	new		
5. Traits - NUE	8. Nitrogen use efficiency (NUE)	continued		
	8. NUE linked to grain quality	new		
	Improvement of water use efficiency and drought tolerance traits	new		
5. Traits - NUE	Resistance to Take-all disease and low soil inoculum build-up	continued		
8. Wheat crossing	11. Introgression of extreme resistance to Septoria tritici leaf blotch from <i>T. monococcum</i>	continued		
9. PCR TILLING		not continued		
	12. Interconnections between the three soil-based explored traits	new		
11. Grain archiving	13. Grain archiving from the diversity and A x C field trials	continued		
6. Sub-contractors	14. Sub-contractor activities	new topics		
	C. The other objectives			
12. Website	15. Website	continued		
13. Electronic Newsletter	16. Electronic Newsletter	continued		
14. The Stakeholders forum and annual conference	17. The Stakeholders forum and annual conference	continued		
16. LINK projects	15. Website	existing and new topics		
	18. Focussed workshops and discussions	new topics		
17. International collaborations	19. International collaborations	continued		
18. Publicity	20. Publicity	continued		

# WGIN Diversity trials 2003 – 2008 at Rothamsted Research:

Nitrogen is a major determinant of yield in crops, however, N-fertiliser application is costly (currently nearing £900/t of N) and excessive application can have negative environmental impacts. Excess nitrogen can affect air quality (via nitrous oxide and ammonia emissions) and highly nitrogenous run-off water from arable land causes major ecological problems including eutrophication of inland lakes, rivers and coastal waters. Improving the use of applied fertiliser will be essential to the success of meeting increasing yield demands for food production whilst limiting damaging environmental impacts.

At the start of this project the extent of genetic variation in traits contributing to the complex issue of efficient use of nitrogen, namely those affecting uptake, conversion into biomass and nitrogen harvest index, within modern wheat varieties was unclear. There had been no comprehensive and systematic analysis of performance in terms of yield and NUE in relation to nitrogen inputs for wheat germplasm. Over the 5 years of the initial project, data for 39 varieties was collected, including a core subset of 15 varieties for which 5 years of data was assembled (see Table 5). The combinations of N treatments, details of application splits and soil mineral N information is shown in Tables 3 and **4**. Wheat was always the first crop following oats. Samples of grain have been archived at -20 °C, and small aliquots (up to 150 g/plot) are available on request.

A range of data was collected (**Table 6**), much of which is available on the WGIN website or is 'in press'. Data included yield and N determinations for grain and straw and all derived NUE parameters. The summarised grain yield data are shown in Figure 2. At all N inputs there exists a range of yield responses when comparing varieties. Independent variation in yield and NUE component performances indicate the possibility of combining these alleles for optimised varieties. In addition, although rankings of performance are often similar at high and low inputs there exist anomalies indicating other novel aenetic variation.

In the new WGIN project, these trials will continue, combining analysis of the core variety subset with inclusion of other novel germplasm. For selected varieties a more detailed analysis of nitrogen partitioning will be undertaken and attention will be paid to post anthesis canopy senescence. In addition, in work supported outside of WGIN, expression of specific gene alleles thought to contribute to NUE will be tested.

For further information on the Diversity Trial, please contact malcolm.hawkesford@bbsrc.ac.uk.

Table 3: I	Table 3: Background information on Diversity trial field design, basic Nitrogen regime and dates								
Harvest year	Drill date	Harvest date	replication	NO <sup>1</sup>	Nitrogen regime Plot size (kg/ha)				Plot size
2004	last week in 11/2003	02/09/2004	3	74	0	50	200	350	10m x 3 m
2005	11/10/2004	11/08/2005	3	30	0	-	200	-	16m x 3m
2006	05/10/2005	09/08/2006	3	86	0	100	200	-	16m x 3m
2007	13/10/2006	30/08/2007	3	52	0	100	200	350	18m x 3m
2008	11/10/2007	19/09/2008	3	59	0	100	200	350	15m x 3m
<sup>1</sup> Soil mineral-N (NO3-N plus NH4-N) to 90 cm depth in February (kg-N/ha)									

Table 4: Diversity trial Nitrogen application regime (kg/ha)								
Total	Splits							
	Mid-March	Mid-March Mid-April Mid-May						
0	-	-	-					
50	50	-	-					
100	50	50	-					
200	50	100	50					
350	50	250	50					

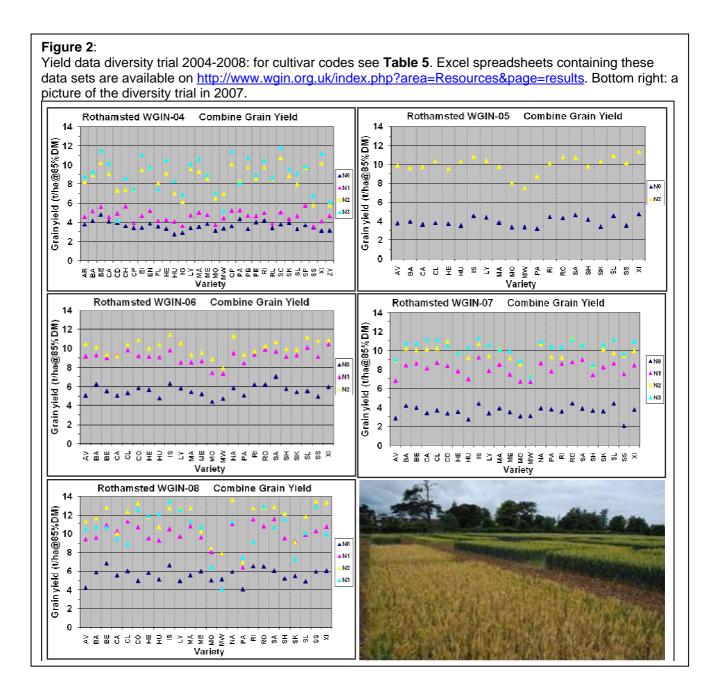
**Table 5**: Varieties used for the Diversity trial in years 2003 – 2008:

			YEAR	OF HAF	RVEST					YEAR	OF HAI	RVEST	TOTAL
NO.	VARIETY	CODE	2004	2005	2006, 2007, 2008	TOTAL NO. OF YEARS	NO.	VARIETY	CODE	2004	2005	2006, 2007, 2008	TOTAL NO. OF YEARS
1	Arche	AR	Х			1	21	Monopol	МО	Х	Х	Х	5
2	Avalon	AV	х	х	х	5	22	Maris Widgeon	MW	х	х	х	5
3	Batis	BA	х	Х	х	5	23	Napier	NA			х	3
4	Beaver	BE	х		х	4	24	Opus	OP	Х			1
5	Cadenza	CA	х	Х	х	5	25	Paragon	PA	Х	Х	х	5
6	Cappelle Desprez	CD	х			1	26	Privileg	PB	х			1
7	Chablis	СН	х			1	27	Petrus	PE	Х			1
8	Claire	CL		Х	х	4	28	Riband	RI	Х	Х	х	5
9	Cordiale	СО			х	3	29	Rialto	RL	Х			1
10	Caphorn	CP	х			1	30	Robigus	RO		Х	х	4
11	Einstein	EI	Х			1	31	Savannah	SA		Х	х	4
12	Enorm	EN	Х			1	32	Scorpion	SC	Х			1
13	Flanders	FL	х			1	33	Shamrock	SH		Х	х	4
14	Hereward	HE	Х	Х	Х	5	34	Sokrates	SK	х	х	Х	5
15	Hurley	HU	Х	Х	х	5	35	Solstice	SL	Х	Х	х	5
16	Isengrain	IG	Х			1	36	Spark	SP	х			1
17	Istabraq	IS		Х	Х	4	37	Soissons	SS	Х	Х	Х	5
18	Lynx	LY	х	х	Х	5	38	Xi 19	ΧI	х	х	х	5
19	Malacca	MA	Х	Х	х	5	39	Zyta	ZY	Х			1
20	Mercia	ME	х		х	4							

Table 6: Field data, harvest data and seed storage information: data was collected in all years marked x

Field data, harvest data and seed storage information	Harvest year				
	2004	2005	2006	2007	2008
Seedling germination measurement <sup>1</sup>	Х				
Soil mineral Nitrogen in February (core depth: 90 cm)	Х	Х	Х	Х	Х
Crop height		x <sup>2</sup>	Х		
Canopy, panicle and leaf temperatures <sup>3</sup>		Х			
Combined grain and straw yield at harvest (t/ha @100% DM)	Х	Х	Х	Х	Х
1000 grain weight				Х	Х
Nitrogen level of grain and straw at harvest	Х	Х	Х	Х	Х
Take-all inoculum build up in soil for a subset of the genotypes <sup>4</sup>	Х	Х	Х	Х	Х
Seven types of derived data <sup>5</sup>	Х	Х	Х	Х	Х
All samples stored at -20C <sup>6</sup>	х	Х	Х	Х	Х

<sup>&</sup>lt;sup>1</sup>Data was collected by Allan Lock, <sup>2</sup> data is available for 200 kg/ha N only, <sup>3</sup>data was collected by Goetz Richter, <sup>4</sup>data was collected by Richard Gutteridge for 200 kg/ha samples, <sup>5</sup>derived data = total yield, harvest index, grain N-uptake, straw N-uptake, total N-uptake, N harvest index and N-Utilisation Efficiency for grain yield; <sup>6</sup> Availability of diversity trial samples: currently 150g/sample maximum; we will consider giving out larger samples at a later date



The replicated diversity trial with large plot sizes and a portion set aside for destructive harvesting permitted researchers to explore several additional traits (**Table 6**). These traits are detailed below.

#### **Germination measurements:**

Allan Lock collected data on germination in the 2003/4 and the 2004/5 seasons. In the 2003/4 season the cultivars Cadenza, Hereward, Malacca, Mercia, Rialto, Soissons and Xi 19 on the N0, N1, N2 and N3 plots were used for measurements. In the 2004/5 season the cultivars were Avalon, Cadenza, Hereward, Malacca, Monopol, Paragon, Shamrock,

Soissons, Solstice, Maris Widgeon and Xi 19 in the N1 and N2 plots.

Mainstem leaf emergence was recorded over the whole season. This was done 17 or 18 times depending on the variety in 2003/4 and 21 or 22 times in 2004/5 on 12 tagged plants which were harvested post flowering. Spikelet number and internode number were recorded and internode length was measured on the surviving mainstems. In addition ear bearing tiller number, but not size was recorded. In 2003/4 width measurements on the final three lamina were done on most plants (some plants had withered in the N0 and N1 samples). In 2004/5 width

WGIN stakeholder newsletter May 2009

measurements were made on all final three leaves of the surviving mainstems.

Most germination data is stored electronically and available to the WGIN community. Allan Lock is currently examining aspects of 'site/ treatment/ fertility' influences on leaf development etc from data that were collected from Broadbalk using identical measurement procedures to those outlined above. This data will be presented at an SEB meeting in Glasgow at the end of June 2009. These examinations are novel and will hopefully provide further insights into the way the plant responds to 'background' and applied Nitrogen. If they prove acceptable they may be applied to the WGIN datasets and may assist with their interpretation. For further information on this dataset contact allan@wicken.u-net.com.

### Canopy, panicle and leaf temperature measurements:

Measurements were taken by Goetz Richter at Rothamsted Research on a clayey silt-loam (Fosters) during the dry summer in 2005 with periods of light heat stress after anthesis. The data series consist of continuous measurements presenting periods of low and high evaporation demand characterised by intermittent rainfall. The potential soil moisture deficit was increasing from about 40 to approximately 180 mm. Weather data were recorded using a Delta-T DL2 weather station in [half-]hourly intervals for all standard variables: rainfall (mm), air temperature (Ta, at 2 m above ground), relative humidity, global radiation and net radiation, and wind speed (m s-1) and direction. The sensors were infrared transducers from Exergen Corporation (Watertown, Massachusetts, USA) of two different types: using a single thermocouple (Ktype; IRt/c.01) and dual thermocouple (EM-type, IRt/c.01. EM). The following varieties were included: Xi 19, Malacca and Solstice, and also on an irrigated reference plot on the Hereward surround. For further information on this dataset contact goetz.richter@bbsrc.ac.uk.

#### **Grain samples requests:**

So far three researchers have requested grain from the diversity trial for further analysis (summarized in Table 7). Peter Shewry and Alison Huttley have acquired two sets of grain samples for the BBSRC CSI project 'Optimising grain shape for improved processing quality'. The project aims to improve the processing quality of UK wheat and barley by optimising grain size and shape, with a particular focus on the development of the grain crease which limits the efficiency of flour production during milling. The major storage tissue in the grain is the endosperm so the project brings together plant scientists with expertise in wheat and barley genetics and cell biology with those studying molecular aspects of endosperm development in maize and Arabidopsis.

Experimental milling is being carried out at the University of Manchester to relate differences in grain architecture to grain mechanical properties and flour yield, with particular emphasis on grain behaviour during the first break. The WGIN samples provide excellent material for this part of the project as we have multiple trials of cultivars with contrasting grain characteristics grown under defined condition.

Alison Huttley's group is interested not only the range of shapes and sizes within the different varieties in the trial but also the effect of nitrogen application. The intention is to measure parameters such as grain length, width and crease depth and carry out microscopic analysis of fixed and embedded material.

#### AxC double haploid trials 2006 - 2008:

The JIC has generated a doubled haploid population from an Avalon x Cadenza cross consisting of 204 independent lines with genetic mapping and phenotype information (see the WGIN website). Within WGIN at Rothamsted we are characterising this population in relation to nitrogen use efficiency (NUE). We aim to map yield, NUE, nitrogen uptake characteristics, nitrogen utilisation efficiency and senescence/nitrogen remobilization rates in this population. Multiple trials will be performed over

Table 7: Diversity trial – use of the grain sample archive (2003 – 2008)								
Samples requested	Experiment	Researcher	E-mail					
aliquots of all 2007 samples	leaf 2 size, leaf 2 N, export of N, gene expression data	Malcolm Hawkesford	malcolm.hawkesford@bbsrc.ac.uk					
selected samples of 2007 harvest	range of shapes and sizes of grain	Alison Huttly	alison.huttly@bbsrc.ac.uk					
10 varieties of 2006- 08 harvests, at selected N levels	relationship between grain architecture and milling properties	Peter Shewry	peter.shewry@bbsrc.ac.uk					

a number of seasons at both high and low

WGIN stakeholder newsletter May 2009

nitrogen inputs. Some preliminary data collected within the first WGIN project indicated extensive variation in these traits and the identification of some putative QTLs. At least one of the yield QTLs appears to be N-dependent. More detailed replicated studies will continue in the new WGIN project and selected lines will be more extensively characterised in terms of N-allocation (at multiple N inputs) and will be coupled with parallel projects investigating the expression of individual key genes involved in NUE. For further information contact malcolm.hawkesford@bbsrc.ac.uk.

#### Avalon x Cadenza Workshop

This is being organised to take place during 2009 to permit all researchers currently working with this mapping population to come together and discuss their findings and future intentions. The workshop is being jointly organised by Simon Griffiths (JIC) and Neil Paveley (ADAS). If you are interested in using the A x C population in the future or your research currently involves its use, please contact us at wgin.defra@bbsrc.ac.uk.

#### **Stakeholders**

Currently, WGIN has 131 registered individuals / organisation representatives who directly receive this newsletter by e. mail. If you know of others who would be interested in the WGIN project objectives or any of the new research topics please could you forward this pdf to them. Our aim is to expand the stakeholder forum to over 300.

#### Website

Elke Anzinger and Pierre Carrion are currently redesigning the WGIN website site to take on the same format as the defra OREGIN project (<a href="http://www.oregin.info">http://www.oregin.info</a>). The new WGIN site layout at the same address (<a href="www.wgin.org.uk">www.wgin.org.uk</a>) will be up and running by early summer 2009. However, from this new site it will still be possible to access all the information on the original WGIN website.

## Final reports available for the WGIN 2003-2008 project

The completed Sid5 and Sid5A forms as well as the detailed 20 page summary of all the results and achieved during the first 5 years of WGIN are now available from the WGIN website.

For further information on the WGIN project please see www.wgin.org.uk or contact us at wgin.defra@bbsrc.ac.uk

The contributors to this newsletter were: At Rothamsted Research: Kim Hammond-Kosack, Malcolm Hawkesford, Peter Barraclough, Peter Shewry, Alison Huttley, Goetz Richter and Elke Anzinger. Allan Lock also contributed.







