

Summary of Progress on the eleven Science Objectives:

Objective 2 Production of Near Isogenic Lines NIL (JIC);

The production of NILs up to BC2 (BC3 equivalent) for height, heading date, and yield QTLs derived from the Avalon x Cadenza segregating population is complete. This activity culminated in a replicated (x3) field trial of the NILs with a total of 1800 entries in 6m² plots in the 2012-13 season. This trial was drilled again in 2013 and the data from both will be used to produce a key WGIN publication for late 2014. One flowering time QTL, located on chromosome 4A, was only identified following the extension of the Avalon x Cadenza linkage map. As a result NIL development is lagging and the lines are not included in the field trial.

In addition to the first cohort of NILs, a set for Lr19, straw wall thickness and grain size have been fixed as NIL homozygotes and are being multiplied from a 2013 drilling in single 1m rows.

Objective 3 The Avalon x Cadenza Mapping population (JIC);

JIC has carried out a round of field multiplication of this population in 2012-13 season and continued to field regular requests for seed supply.

Objective 4 Paragon gamma and EMS mutagenised populations (JIC);

Both populations are maintained at JIC. DNA was extracted from 2500 of the gamma deletion lines and aliquots have been supplied to users of this resource. The materials form an important element of several funded projects and publications.

Objective 5 AE Watkins and Gediflux collections (JIC);

A manuscript describing the genetic structure of the Watkins and Gediflux collections was submitted for publication. Seed for both collections is widely accessed by users and this is handled through WGIN and BBSRC prebreeding LOLA.

Objective 6 New mapping populations (JIC)

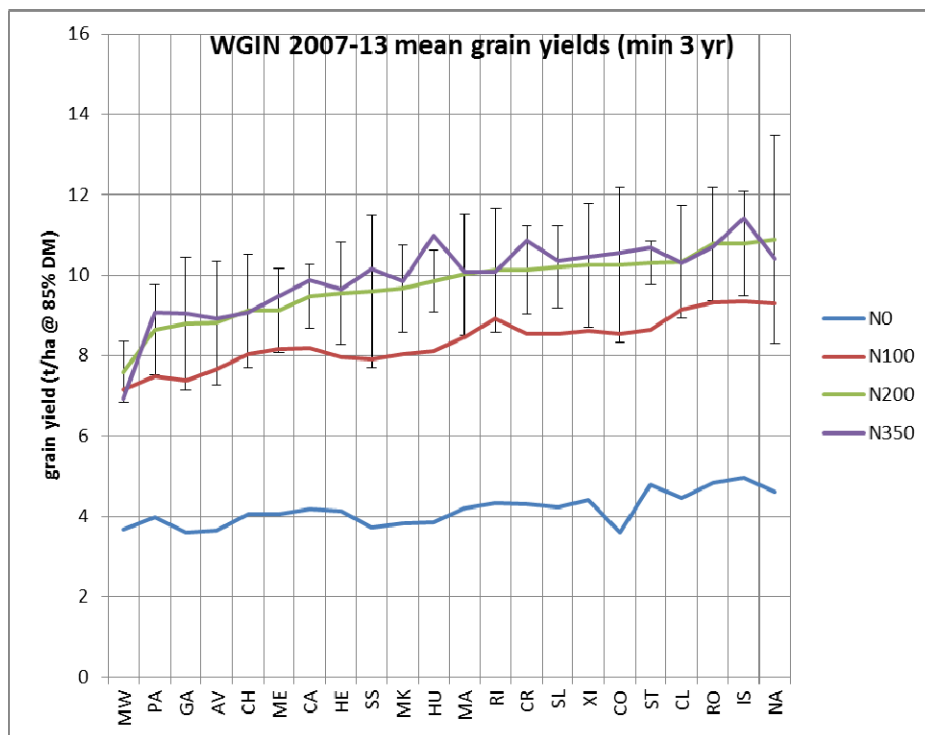
The Paragon x Garcia, Paragon x Chinese Spring, and Paragon x Synthetic single seed descent populations are complete. The BBSRC CiRC project 'Development and validation of a flexible genotyping platform for wheat' developed high resolution genetic maps for these populations, which like the germplasm are fully available on the WGIN and Cerealsdb website.

Objective 7 Insect resistance in wheat: Cereal aphids (RRes) – no funded research activities beyond year 1

Objective 8. Nitrogen use efficiency (NUE) and Quality QTLs linked to NUE (RRes)

As in previous years, a field trial (The Diversity Trial) examining interactions of NUE and genotype was drilled in the autumn of 2012 and harvested in August 2013. Grain and straw yields were recorded and samples taken for nitrogen analysis. This trial was made available to the community for phenotyping/sub sampling.

This trial examined 25 varieties subject to 4 levels of N fertiliser application (0, 100, 200 and 350 kg/ha) in a triplicate design. The 2013-harvest varieties consisted of 25 elite current or older varieties. New varieties for 2013 harvest are AC Barrie (chosen because of exceptional high protein content and quality) and Cocoon (a taller variety which may have high nitrogen acquisition and utilisation capacity). These replace two of the previously selected Avalon x Cadenza DH lines.



The figure shows mean yields of varieties with at least 3 years' data since 2007, for each of the N levels, ranked on performance at 200 kgN/ha (N200). Evident is the large impact of adding N fertiliser (compare N0 with N100), the relatively modest but significant yield increase between N100 and N200, and the minimal response above that (see N350). It is also clear that the rank order is comparable at all N input levels. Standard deviations are also plotted for N200, clearly indicating the considerable year to year variation over the period of the study.

The second figure in this section shows mean grain N concentration of the 33 varieties for which there is at least 2 years data since 2004 for N200 (200 kgN/ha). The varieties are ranked on their mean performance.

conductance, that the increased grain yield under drought amongst cultivars (Fig. 9.2) was associated with deeper roots.

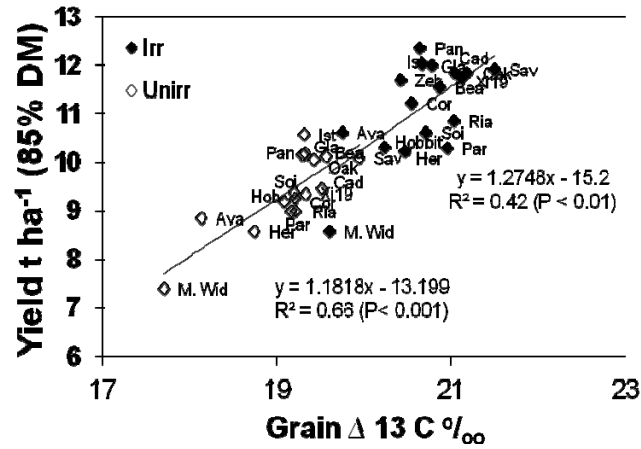


Fig. 9.1a Relationship between grain $\Delta^{13}C$ vs grain yield in 18 wheat cultivars for Sutton Bonington (mean Sutton Bonington 2009-10 and 2010-11) under irrigated and unirrigated conditions.

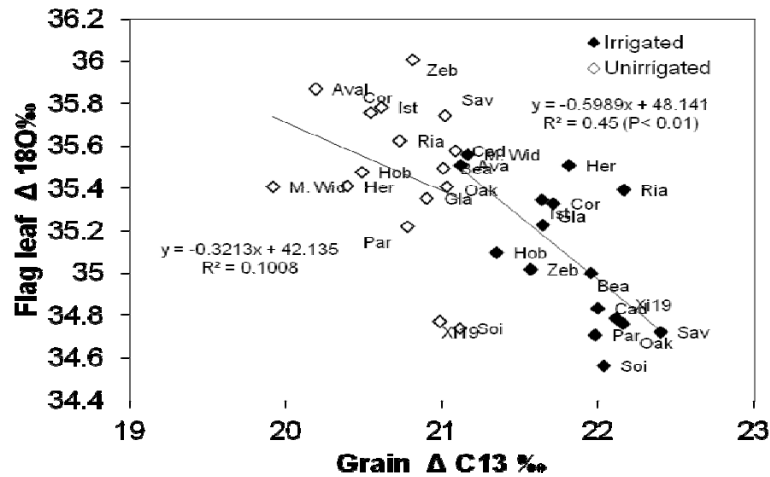


Fig. 9.1b Relationship between grain $\Delta^{13}C$ and flag leaf $\Delta^{18}O$ in 18 wheat cultivars (mean Sutton Bonington 2009-10 and 2010-11) under irrigated and unirrigated conditions.

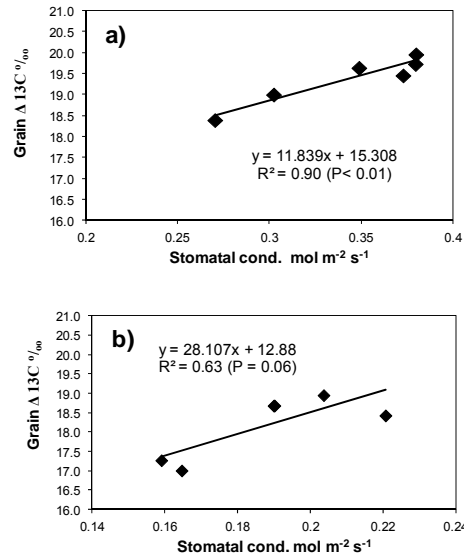


Fig. 9.2 Relationship between grain $\Delta^{13}C$ and stomatal conductance in 18 wheat cultivars (mean Sutton Bonington 2009-10 and 2010-11) under irrigated and unirrigated conditions.

Objective 9.2 “To identify robust QTLs for water-use efficiency”

One DH mapping population (Rialto x Savannah, 94 lines) has been phenotyped at Nottingham University (2 reps) under irrigated and un-irrigated conditions in 2012-13. In all sub-plots, anthesis date, plant height, combine grain yield and yield components were measured. Additional measurements in un-irrigated plots included: (i) % stem water soluble carbohydrate at GS61+10d, (ii) leaf senescence kinetics for flag-leaf, L2 and L3 at 3-4 day intervals post-anthesis and (iii) grain $\Delta^{13}C$. Overall the grain yields in the un-irrigated plots (7.66 t/ha) were significantly decreased compared to the irrigated treatment (10.36 t/ha) t ha⁻¹ ($P = 0.06$). There was a statistically significant interaction between irrigation and DH line ($P < 0.001$; Fig. 9.3).

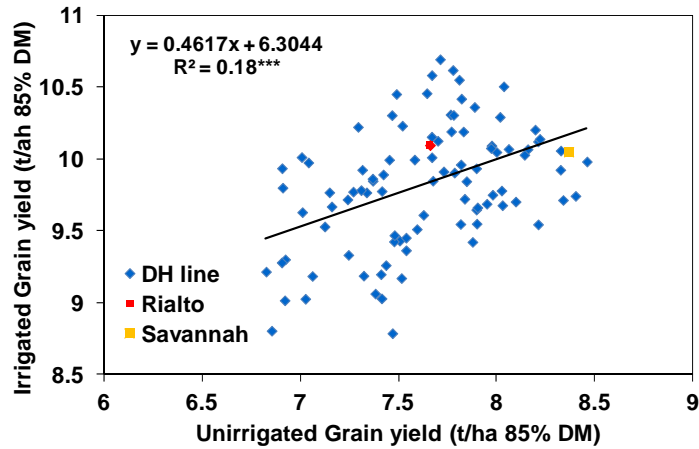


Fig. 9.3 Grain yield in irrigated and unirrigated plots at Sutton Bonington in 2012-13 ; LSD (5%) Irr = 2.97 (P = 0.06); Line 0.51 (P< 0.001) and Irr x Line = 0.95 (P< 0.01).

Although there were statically significant differences between stem water soluble carbohydrate (WSC) accumulated at GS61+9 days in the rage 209-512 g m⁻² (P< 0.001). there was no association between stem WSC and grain yield under drought (data not show). Senescence traits measured in the un-irrigated treatment were, however, associated with grain yield performance amongst the 94 DH lines, e.g. the Normalized Difference Vegetation Index (NDVI) measured during mid-grain filling on 16 July (Fig. 9.4).

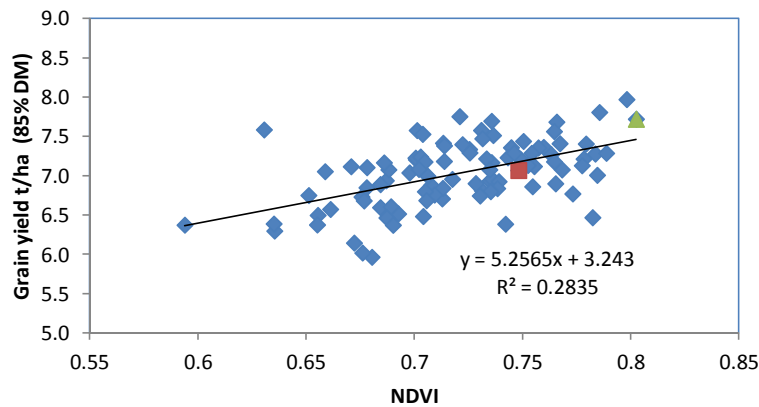


Fig. 9.3 Grain yield versus NDVI on 16 July in unirrigated plots at Sutton Bonington in 2012-13.

Data from the 2010-11 experiments for Rialto x Savannah DH population at Nottingham (irrigated and unirrigated) and John Innes Centre (unirrigated) were submitted in January 2013 for QTL analysis using an existing molecular marker map at JIC for this population (anthesis date, plant height, combine grain yield and yield components, Δ 13C and senescence parameters; Table 9.5).

Table 9.1 QTL analysis for traits in Savannah x Rialto DH population at Nottingham in 2010-11

chr	pos	LOD	var	mean	add eff	on	marker	trait	env
1D	28	3.1	12.9	0.5	-0.008	S	RAC875_c78062	HI	IRRI11
4D	17.8	2.7	11.1	0.5	0.008	R	GENE-2812	HI	IRRI11
4A	191.6	2.5	7.7	50.8	-0.8	S	BS00023164	TGRWT	IRRI11
5A	70.8	5.8	20.1	50.8	-1.35	S	Kukri_c61108	TGRWT	IRRI11
6A	129	4	13.1	50.8	-1.1	S	EXcalibur_c52196	TGRWT	IRRI11
2D	3	3	12	343	12.9	R	Kukri_c2912	EARNBpsqm	UNIRRI11
7A	0	3.9	16	343	15.1	R	EXcalibur_c48636	EARNBpsqm	UNIRRI11
2D	3	4.4	17.3	16267	703	R	Kukri_c2912	GRpsqm	UNIRRI11
3B	261	3	11.3	16266	-585	S	Ex_c14162_22093694	GRpsqm	UNIRRI11
3B	260	2.4	11.3	7.6	-0.19	S	Ex_c14162_22093694	GRYLD	UNIRRI11
3A	144.9	2.6	7.2	47	1.2	R	BS00110564	TGRWT	UNIRRI11
5A	337.3	2.8	7.7	47	-1.3	S	IAAV7514	TGRWT	UNIRRI11
7D	45	2.9	8.1	47.2	-1.1	S	Kukri_c48125	TGRWT	UNIRRI11
2D	34	8.2	30.3	4.4	0.28	R	BS00049370	LFCURL	UNIRRI11
2B		3.0	17.5	4.4	0.18	R	BS00098024	LFCURL	UNIRRI11
5A	119.8	1.4	4.2	4.4	0.18	R	RAC875_c61493	LFCURL	UNIRRI11
5A	249.8	3.2	15.1	0.32	-0.011	S	EXcalibur_rep_c68005	NDVI	UNIRRI11
2A	161	3.4	16.0	18.2	0.135	R	Tdurum_contig66015	DeltaC	UNIRRI11

In un-irrigated conditions, a QTL for grain Δ^{13} (DeltaC) was found on chr 2A. In both irrigated and un-irrigated conditions, a QTL for thousand grain weight (TGRNWT) identified on chr 5A. In irrigated conditions, a QTL was found for harvest index (HI) on chr 1D and 4D and for TGRNWT on chr 4A and 6A. In addition, in un-irrigated conditions, significant QTLs were located for grains m^{-2} (GRpsqm) on 2D and 3B, grain yield (GRYLD) on 3B, and leaf curling on 2B, 2D and 5A. Grain samples were sent to the laboratory for Δ^{13} determination in December 2013 and the complete phenotyping dataset for the 2012-13 experiment will be sent to JIC for QTL analysis in January 2014.

Objective 9.3 “To develop one DH population for drought-tolerance research”

See JIC report above **Objective 6**.

Objective 9.4. “To identify novel alleles for the AE Watkins and Gediflux collections”

At Nottingham University, assessments of: (i) flag-leaf chlorophyll fluorescence (PSI Fluorpen) and (ii) canopy NDVI were carried out on 96 AE Watkins lines in a field experiment on a sandy loam site during grain filling in 2010-11. Significant variation was identified amongst lines in the stay green trait (as indicated by NDVI) and flag leaf photosynthetic rate (as indicated by chlorophyll fluorescence quantum yield). For both traits there were significant correlations with grain yield amongst lines indicating the value of these traits a selection criteria for improving drought tolerance (Table 9.2).

Table 9.2. Maximum, minimum and mean values of Normalized Difference Vegetation Index (NDVI) and flag-leaf chlorophyll fluorescence quantum yield (QY) for Watkins landrace lines at Nottingham in 2010-11.

	Grn Yield t/ha	<u>Normalized Difference Vegetation Index</u>					<u>Quantum Yield</u>		
		03-Jun	14-Jun	23-Jun	13-Jul	22-Jul	03-Jun	13-Jun	22-Jun
Max	714.7	0.86	0.87	0.86	0.75	0.49	0.72	0.71	0.73
Min	221.4	0.57	0.64	0.59	0.33	0.22	0.46	0.48	0.49
Mean	495.3	0.74	0.76	0.73	0.53	0.30	0.63	0.62	0.65
LSD	159.28	0.066	0.072	0.032	0.069	0.052	0.117	0.115	0.0891
Prob.	< 0.001	<0.001	<0.001	< 0.001	< 0.001	< 0.001	< 0.01	< 0.05	< 0.001
Correl G Yd		0.58	0.63	0.62	0.54	0.30	0.15	0.035	0.16
Prob. Correl		<0.001	<0.001	<0.001	<0.001	<0.001	0.098	0.698	< 0.001

Objective 9.5. “Collate a diverse germplasm (cvs, advanced lines) for future association genetics studies”

Seed of set of 60 spring wheat cultivars and advances lines (the CIMMYT Core Germplasm 1 CIMCOG panel, Wheat Yield Network) has been provided by CIMMYT, Mexico under an appropriate MTA to Nottingham University, Rothamsted Research and John Innes Centre for future drought association genetic studies and other relevant research activities .

Objective 10. Resistance to take-all disease (RRes)

Objective 10.1. To identify wheat germplasm resistant to take-all.

Screening of the Watkins and Gediflux collection for resistance to the take-all fungus has been carried out over 5 years. Combing the results from previous years the final year of screening was carried out in 2012 for the remaining 104 Watkins lines and 36 Gediflux lines. These samples are currently being assessed for take-all disease. Results from other trials carried out in 2012 show that it was a very high take-all disease pressure year. This should allow good discrimination between the lines tested and identify any final Watkins and Gediflux lines showing resistance to take-all disease over multiple field trial years. From previous WGIN Watkins/Gediflux field trials the most resistant 144 Watkins lines and 12 Gediflux lines have already been taken forward in the replicated wheat pre-breeding Lola field trials. If any more lines from the 2012 WGIN Watkins/Gediflux field trial are identified as highly resistant these will also be taken forward into further years of replicated trials in the pre-breeding Lola.

Objective 10.2. Genetic analysis of resistance to take-all in hexaploid and non-hexaploid wheats.

The F₆ mapping population between *Triticum monococcum* lines MDR037 (susceptible to take-all disease) x MDR046 (resistant), developed during the WGIN programme, has been sown in a third wheat replicated field trial in November 2013.

Plant samples will be taken in summer 2014 to be assessed for take-all disease to identify the genetic loci conferring resistance.

Two other *Triticum monococcum* mapping populations (MDR031 x MDR043 and MDR043 x MDR046) are currently at the F₃ stage.

Objective 10.3. Introgress resistance to take-all from different non-hexaploid wheats.

The introgression crossing programme involving the 5 selected take-all resistant *T. monococcum* lines (MDR031, MDR046, MDR229, MDR232 and MDR286) is now in progress. All five lines have been successfully crossed to the Paragon *ph1* mutant. The lines will now be backcrossed into Paragon without the pairing locus mutation.

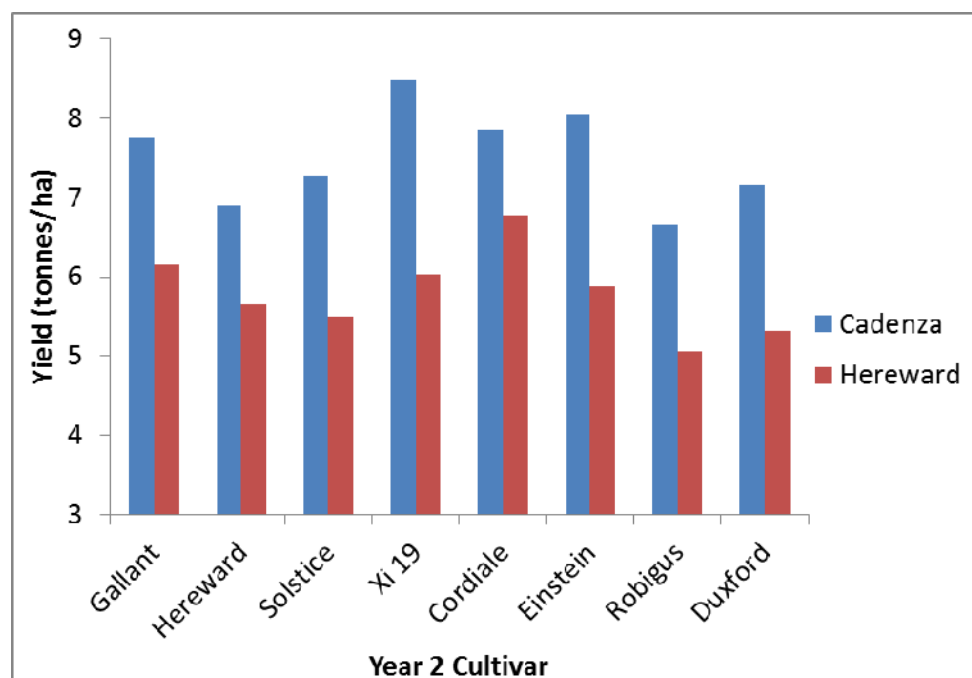
Objective 10.4. Identification and characterisation of hexaploid wheat germplasm which reduce take-all inoculum build-up (TAB) in soil.

After harvest in 2013 the WGIN Diversity trial was sampled to phenotype all 25 cultivars for the TAB trait. These cultivars were sampled at the 200 kg/ha N rate. A subset of cultivars was also sampled at the 0, 100 and 350 kg/ha N rates to investigate the interaction between nitrogen rate and take-all inoculum build-up. These were the low TAB cultivars Cadenza, Cordiale and Xi 19, the high TAB cultivars Avalon and Hereward, and the intermediate cultivar Riband. These samples will be processed in spring 2014.

Within WGIN 2 cultivar rotation trials were set up to explore the significance of the TAB trait on the amount of take-all disease and yield in a following second wheat. In the first year these trials consisted of large 82 m x 12 m strips of the low TAB cultivar Cadenza and high TAB cultivar Hereward (4 replicates of each cultivar). In the second year these strips were divided into 8 smaller plots (10 m x 3 m) and sown with 8 randomised winter wheat cultivars (Cordiale, Duxford, Einstein, Gallant, Hereward, Robigus, Solstice, Xi 19). After harvest in the first year five soil cores were taken from each of the designated Year 2 plots to gauge the amount of take-all inoculum build-up. In the second year plant samples taken in spring and summer for take-all disease assessment and yields were recorded at grain maturity.

The final cultivar rotation trial has now finished its second year (2012-2013). In the first year there was, as expected, a significantly higher ($P = 0.019$) build-up of take-all inoculum under Hereward (37.6% roots infected in soil bioassay) than Cadenza (12.8%). In spring of the second year this resulted in a much higher incidence of disease for the eight winter wheat cultivars sown after Hereward compared to Cadenza in the first year (average 78.9% plants infected in the second wheat sown after Hereward, 27.6% after Cadenza, $P < .001$). Plant samples were taken in the summer at GS 77. These were washed and dried for storage and disease assessment over the winter. These take-all assessments are currently in progress. Yields taken at grain maturity have revealed a significant yield advantage in the second year when the eight wheat cultivars were sown after Cadenza compared to

Hereward (Figure 10.4, $P = 0.004$). This was independent of second wheat cultivar choice ($P = 0.737$) and demonstrates the potential benefit of the low TAB trait to reduce the amount of take-all disease and improve second wheat yields. A manuscript is currently being prepared on the WGIN cultivar rotation trials.



Objective 10.5. To explore the genetic basis of take-all inoculum build-up.

The 2012 WGIN AxC experiment (204 AxC lines + parents, 3 reps) (12/R/WW/1202) in the Rothamsted field Bones Close was sampled for the TAB trait after harvest in September 2012. These samples were assessed in 2013 and a QTL analysis will be carried out over the winter. The aim is to confirm the location of the two putative QTLs conferring the low TAB trait in Cadenza.

Objective 11. Introgression of extreme resistance to Septoria leaf blotch from *Triticum monococcum* into hexaploid wheat (RRes)

The introgression crossing programme involving the 5 selected take-all resistant *T. monococcum* lines (MDR236, MDR308, MDR1959, MDR1976 and MDR2055) is now in progress. All five lines have been successfully crossed to the Paragon *ph1* mutant. The lines will now be backcrossed into Paragon without the pairing locus mutation.

Objective 12. Interconnections between the three soil-based explored traits (RRes and UoN)

The final data sets were obtained in the 2013 field season. The data obtained over the 5 years on NUE components, drought tolerance components and resistance to take-all disease and take-all inoculum build up will now be inter-compared in the 4 month WGIN extension.