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SID 4

Report for Period 2009

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Scientific objectives

7. Please list the scientific objectives as set out in the contract. If necessary these can be expressed in an abbreviated form. Indicate where amendments have been agreed with the Defra Project Manager, giving the date of amendment.

01 Project management (RRes); 02 Production of Near Isogenic lines (JIC); 03 The Avalon x Cadenza Mapping population (JIC); 04 Paragon gamma and EMS mutagenised populations (JIC); 05 AE Watkins and Gediflux collections (JIC); 06 New mapping populations to align WGIN 2 with the International wheat genome sequencing (JIC); 07 Insect Resistance (RRes); 08. Nitrogen use efficiency (NUE) and Quality QTLs linked to NUE (RRes); 09 Drought tolerance (University of Nottingham); 10 Take-all disease (RRes); 11 Introgression of extreme resistance to Septoria leaf blotch from Triticum monococcum into hexaploid wheat (RRes);12 Exploring the Interconnections between the three soil based traits; 13 Grain archiving; 14. Sub-contractor projects (To be determined during the project) 15. Website; 16 Electronic Newsletter; 17 Annual Stakeholders Forum; 18 Focussed workshops; 19 International collaborations; 20 Publicity

Summary of Progress

8. Please summarise, in layperson's terms, scientific progress since the last report/start of the project and how this relates to the objectives. Please provide information on actual results where possible rather than merely a description of activities.

Objective 1. Two management meetings were held in March (X), June (X) and October (U of Nottingham). The new style of open morning meeting and focused milestones afternoon meeting with just two breeder representatives has worked well. All minutes and ppts have been posted on the WGIN website within 3 weeks of each meeting taking place.

The progress on the 11 wheat research objectives (2 through 13) is summarised in an accompanying word document. Overall, additional genetic and phenotypic variation for each of the traits of interest has been identified and increased precision has been made through the generation of near isogenic lines and/or the establishment of closely linked markers or new mapping populations. The large number of field experiments planned for year 1 were completed and the required grain archived (objective 13). In addition the seed stocks generated at RRes can now be accessed via the JIC - Genetic Resources Unit website.

Objective 14 The new sub-contracting procurement procedure was implemented. In total 5 applications were received and are currently being assessed by external reviewers.

Objectives 15, 16 and 17. The WGIN website was completely overhauled by mid-summer 2009, two newsletters appeared on time (May and October), and the stakeholders meeting at RRes was very well attended in Nov with 25% new faces amongst the audience of ~100. This was helped by us preparing an article for the Next Generations Foods website which appeared in late Sept and by using six organisations mailing lists to advertise this event to ~ 2000 individuals primarily based in the UK. Representatives from TSB, KTN and BBSRC gave ppts explaining the forthcoming new funding opportunities for wheat. The newsletters and stakeholder ppts are available from the website.

Objective 18. The Avalon x Cadenza workshop was held at JIC in Nov with ~ 60 UK researchers attending. There are at least 18 traits being actively explored in the public domain using this mapping population as well as several others in the private domain.

Objective 19. Our intention is to link up with wheat breeders from the far east of Europe with a Joint workshop in Serbia planned for early summer 2010. A BBSRC ISIS application has been submitted to support the travel of 12 UK wheat scientists. The successful outcome was known in Jan 2010.

Objective 20. The scientific outreach activities were numerous, with displays by both JIC and RRes staff on specific WGIN objectives at Cereals 2009, via field demonstrations and talks at the respective sites and engaging the media.

Amendments to project



Progress in relation to targets

10. (a) List the agreed milestones for the year/period under report as set out in the contract or any agreed contract variation.

It is the responsibility of the contractor to **check fully that all milestones have been met** and to provide a detailed explanation when they have not been achieved.

Milestone			Milestones met		
Number	Title	Target date	In full	On time	
01	Project management	30/11/2013		 ✓ 	
02	Production of Near Isogenic Lines	01/03/2013		V	
03	The Avalon x Cadenza mapping population	30/11/2013		V	
04	Paragon gamma and EMS mutagenised populations	01/01/2011		\checkmark	
05	AE Watkins and Gediflux collections	01/07/2011		no	
06	New mapping populations	01/03/2013		\checkmark	

07	Insect Resistance	01/04/2010	\checkmark
08	Nitrogen use efficiency (NUE) and Quality QTLs linked to NUE	30/11/2013	\checkmark
09.1	Drought tolerance: Complete phenotyping and data analysis for drought tolerance traits in elite winter wheat varieties in 2009/10 &10/11	30/11/2011	✓
O9.2	Drought tolerance: QTL analysis to identify genome locations associated with WUE and drought tolerance traits completed.	30/11/2012	\checkmark
O9.3	Drought tolerance: Complete development of one new DH population in an elite modern background segregating for drought-tolerance traits.	31/03.2012	\checkmark
O9.4	Drought tolerance: Association genetics analysis of drought tolerance traits using AE Watkins & Gediflux collections completed.	28/02.2013	\checkmark
O9.5	Drought tolerance: Collation of diverse germplasm collection (cultivars, advanced lines) from worldwide drought-tolerance wheat breeding programmes completed.	28/02/2013	
10	Take-all disease	30/11/2013	✓
11	Introgression of extreme resistance to Septoria leaf blotch from Triticum monococcum into hexaploid wheat	30/11/2012	no
12	Exploring the Interconnections between the three soil based traits	30/11/2013	Ý
13	Grain archiving	30/11/2013	~
14	Sub-contractor projects	30/11/2013	✓
15	Website	30/11/2013	~
16	Electonic Newsletter	30/11/2013	✓
17	Annual Stakeholders Forum	30/11/2013	
18	Focused workshops	30/11/2009 ✓	
19	International collaborations	30/11/2013	
20	Publicity	30/11/2013	\checkmark

For objective 5, some of the Watkins collections lines have so far failed to produce sufficient seed. Also some lines revealed two different phenotypes when grown in the field in 2009 and so are now being multiplied as separate lines.

For objective 11, all genotyping and phenotyping were delayed due to a change in personnel in June 2009 and the replacement not arriving at Rothamsted till Nov 2009. Dr Hai-Chun Jing who was an intergral part of the WGIN project since March 2004 left Rothamsted in July to become a programme leader at the Centre for Bioenergy Plants Research and Development, Institute of Botany, Chinese Academy of Sciences, 20 Nanxincun, Xiangshan, Beijing 100093 to develop research into abiotic stress in sorghum. Temporal hire Steve Freeman completed some of the required activities. New permanent hire Dr Wing Sham Lee trained at Cambridge University will now be trained by Kostya Kanyuka and Kim Hammond-Kosack to achieve these objectives.



11. (a) Please give details of any outputs, e.g. published papers/presentations, meetings attended during this reporting period.

Peer Reviewed:

Simon Griffiths, James Simmonds, Michelle Leverington, Yingkun Wang, Lesley Fish, Liz Sayers, Leodie Alibert, Simon Orford, Luzie Wingen, Laurence Herry, Sebastien Faure, David Laurie, Lorelei Bilham, John Snape. Meta-QTL analysis of the genetic control of ear emergence in elite European winter wheat germplasm. Theoretical and Applied Genetics 119 (2009), <u>doi 10.1007/s00122-009-1046-x</u>

Jing HC, Bayon C, Kanyuka K, Berry S, Wenzel P, Huttner E, Kilian A, Hamond-Kosack KE. (2009) DArT markers: diversity analyses, genomes comparison, mapping and integration with SSR markers in Triticum monococcum. BMC Genomics, 10: 458. <u>doi:10.1186/1471-2164-10-458</u>

Non-Peer reviewed:

Popular press articles:

16.06.2009: Article in Farmers Weekly on wheat take-all (<u>http://www.fwi.co.uk/Articles/2009/06/16/116155/Wheat-take-all-research.htm</u>) August 2009: The diversity trials were filmed as part of the BBC report 'Averting a perfect storm of shortages' (<u>http://news.bbc.co.uk/1/hi/world/8219184.stm</u>)

Article written for the Next Generations Foods website entitled WGIN - Improving the Environmental Footprint of Farming available at http://www.nextgenerationfood.com/article/WGIN

The WGIN T. monococcum field trial appeared in the BBSRC brochure 'The Bioscience behind: Secure harvests' in the section 'Getting novel traits into wheat'.

Oral Scientific Presentations:

Project promoted in School of Biosciences Open Day, University of Nottingham 23 June

BSc undergraduate and MSc Plant and Crop Sciences students from the University of Nottingham (Feb)

Scientific Discussions:

A joint wheat discussion meeting involving several representatives from Cimmyt, BBSRC, and the Bill and Melinda Gates Foundation took place on 12th Dec 2009. Present from the WGIN project were Peter Shewry, Simon Griffiths and Kim Hammond-Kosack.

Poster Scientific Presentation:

At Cereals 2009 and the WGIN stakeholder meeting in Nov and the Monogram meeting at the University of Bristol in April

Scientific Outreach activities / articles:

WGIN was represented at Cereals 2009 at the stands of JIC and RRes and Nottingham. Presentations by Richard Gutteridge (RREs) (1) At 3 HGCA Roadshow. Take-all - Predicting the risk, includes work within WGIN on varieties. (Peterborough. Newbury and Willerby, Jan). (2) To AICC (Eastern region) and farmers on Strategies for take-all control, includes work within WGIN on wheat varieties. (Duxford. Feb). (3) Take-all workshop (Brooms Barn, March) and (4) Writtle college students (Nov).

Field demonstration of the WGIN experiments at RRes to Writtle college students (April), Country Land Association - Hertfordshire and Middlesex branch (May), St.Alban's Young Farmers (May), Dutch cereal pathologists (May), Syngenta trainee agronomists (June), South Bedfordshire farmer's group (June), German farmers (June), HGCA R & D committee, Syngenta and BBSRC (June), Defra science advisory council (14th July) and Savills agronomists (July).

Property a	ortunities for explo arising out of this w ease give details.	iting Intellectual ork been identifie	ed?		 YES 🗌 I	NO
	ject is entirely IP fro	ee			 	
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Has any o If YES , ple	other action been ta ease give details.	aken to initiate Kr	nowledge Trar	nsfer?	 YES 🔀 I	NO

Two of the biotic stress objectives topics are exploring the possibility of using the new Technology Strategy Board (TSB) initiative call in crop projection, to engage relevant industries to exploit further the knowledge gained from the WGIN project.

Future work

12. Please comment briefly on any new scientific opportunities which may arise from the project.

Objective 9 (Drought tolerance): It is too early in the project to identify new scientific opportunities with reasonable certainty.

Objectives 2, 3, 4, 5, 6, 7, 8, 9, 10 and 11 additional research activities have been proposed within a new proposal submitted to BBSRC within the wheat pre-breeding LOLA lead by Graham Moore at JIC as themed pillars of research.

Objective 10 is being taken forward as a BBSRC strategy LOLA application on Take-all involving JIC , NIAB and RRes lead by Anne Osbourn at JIC. HGCA funds are also being sort to support this project.

Declaration

13. I declare that the information I have given is correct to the best of my knowledge and belief.

Name

Date

Position held

Summary of Progress on the eleven Science Objectives:

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

<u>Avalon x Cadenza NILs</u>: At the selfing stage following second backcross for 3A height (into Avalon and Cadenza). The NIL are close to completion for this set.

Completed second backcross for 1B heading / late, 1D heading early, 2A height / short, 2D height / tall, 2D height yield, 3B height / short, 5A yield, 6A height, 6B height, 7B and 7D yield (all to Avalon). 1B heading early, 1D heading, 2D height, 3B yield, 2A height / tall, 2D height, 6A height, 6B height and heading, 3B height (all to Cadenza). Self stage to follow

Completed third backcross for 2DL, 7DL straw wall thickness (into Avalon). Self stage to follow.

Completed F1 production for 3BL straw wall thickness. Marker Assisted Selection (MAS) to begin at next stage for these (into Avalon and Cadenza) production for 3BL straw wall thickness. MAS to begin at next stage for these.

<u>Rialto x Savannah NIL</u>: Completed F1 production for 3D stay green / late and early, 7D stay green / late and early (into Rialto, Savannah and Paragon). MAS to begin at next stage for these.

<u>Lr19 NILs</u>: Leaf rust resistance gene Lr19 from *Agropyron elongatum* is being followed in 2 sets of crosses, Oasis, Kamb1 and Wheatear x Paragon and Oasis, Kamb1 and Wheatear x Alchemy. Completed first backcross analysis using the Lr19 specific marker.

<u>Malacca x Hereward NILs</u>: A range of bread-making quality traits and measurements including loaf volume and cell number are being followed in this cross. Completed first backcross using MAS.

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

A new version of the Avalon x Cadenza map has been released for use on the WGIN website. The JIC map has been created for general use as a pre-made tool using "Joinmap". It comprises 28 linkage groups, covering 2000cMs. A mixture of marker types (SSRs, DaRT, COS, AFLP and perfect markers) has been incorporated to give the best overall coverage, and co-segregating markers have been kept to a minimum. This map gives a good overview of the cross, and would be suitable for QTL analysis. Quality control on this map has been high, with all data checked, and the map order of the markers verified using a new web based mapping tool called Threadmapper (<u>http://cbr.jic.ac.uk/threadmapper</u>). All mapping data is available so that it is possible to create your own map. This may be useful for cross referencing purposes.

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

<u>Gamma:</u> 450 M3 lines (250Gy) of Paragon developed with DNA extracted 1000 M2 lines of Paragon developed with 4000 more M1 seeds available (all 250Gy)

<u>EMS</u>: 6500 M6 lines developed from field trials. Populations developed from mutants (heading late, height short, stay green early / late) to F3 with spring cultivars and sown to field trials autumn 2010 for het selection

Yield and stay green mutants that have been identified are currently under field tests with the breeding company RAGT seeds

Objective 5 AE Watkins and Gediflux collections (JIC);

<u>AE Watkins</u>: 1060 Watkins lines have been sown in autumn 2009 to 1m plots for bulking to a minimum of 200g and trait recording in 2010. The number of lines reflects the original 814 accessions plus extra diversity discovered within accessions.

The same 1060 lines are being grown as single plants under glass for precise genetic stocks (bagged seed) and DNA has been extracted from these.

All lines were photographed before sowing to complement seed size data recorded in summer 2009 on MARVIN analyser.

Ten populations of SSD Watkins x Paragon to F3 seed (heading late / early, height tall / short). Material sown for F1 production of further populations (seed size large / small)

<u>Gediflux:</u> 64 key diverse lines of the collection bulked to a minimum of 200g. Remaining 440 lines to be brought up to this quantity summer 2010.

Objective 6 Developing two new Mapping populations: Not yet started

Objective 7 Insect resistance in wheat: Cereal aphids (RRes) (note - this trait was only funded to be studied in year 1)

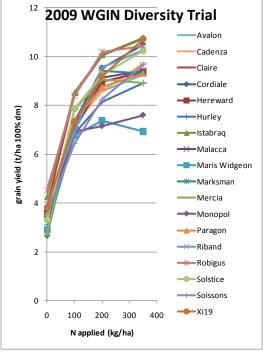
7.1.: To explore whether the differential response of hexaploid wheats to two different cereal aphid species has a genetic basis:

Milestone 1. Determining the differential susceptibility to two cereal aphid species of targeted lines from the Spark x Rialto mapping population.

In previous studies, some lines from the Spark x Rialto mapping population showed strong resistance or susceptibility to Russian wheat aphid, Diuraphis noxia and greenbug Schizaphis graminum, but only one line had a consistent effect against both aphid species. Seventeen of these extreme lines, plus the parents, have now been tested against UK cereal aphids Rhopalosiphum padi and Sitobion avenae in laboratory bioassays. Replicated groups of 10 alate aphids were given the choice between two seedlings, at the first leaf growth stage, one of a standard variety, Solstice and the other of the test variety. The number of alates settled and the number of nymphs produced on each seedling was recorded at 24h. These data were then compared in a paired Students t test and the number of nymphs produced on the test line was expressed as a proportion of the nymphs produced on Solstice in the same assay, thus providing a "preference index" for both aphid species. As for D. noxia and S. graminum, the most and least preferred lines were not the same for R. padi and S. avenae. However, there were a few lines where the responses coincided and one in particular that was very susceptible to 3 of the 4 aphid species. These data are now being checked to determine whether there is any genetic basis to the effects and the lines of extreme preference for R. padi and S. avenae are being tested in aphid development assays to confirm the results. In addition, the possible role played by a family of plant defence secondary metabolites, the benzoxazinones e.g. DIMBOA, in the contrasting preference index of the two aphid species for these lines will now be explored.

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

Two field trials were planted in the autumn of 2008 and harvested in August 2009. In both cases grain and straw yields were recorded and samples taken for nitrogen analysis (in progress). In addition canopy spectral reflectance data were collected to assess post anthesis canopy longevity. These trials were made available to the community for phenotyping/sub sampling. The first of these trials (The Diversity trial) examined 24 varieties subject to 4 levels of N fertiliser application (0, 100, 200 and 350 kg/ha) in a triplicate design. The varieties consisted of 18 elite current or older varieties (see figure showing yield response) and 6 lines selected double haploid lines from the mapping population (Avalon x Cadenza). The selected double haploid



lines (chosen for contrasting NUtE, leaf N and timing of senescence) were subject to a more detailed yield component analysis following anthesis and at final harvest. The second trial (The Avalon x Cadenza Mapping Population Trial) consisted of all 204 lines of the mapping population, grown in triplicate at 100 kg N/ha. Combining these data will enable calculation uptake ability and nitrogen of nitrogen utilisation efficiency (yield per unit N taken up). In addition the trials were phenotyped and sub sampled for a number of separately funded projects including assessments of grain protein quality, transcriptome, canopy structure and senescence, and nitrogen content at and just after anthesis.

The phenotyping data (yield, NUE parameters, canopy senescence) being collected on the mapping population is being

averaged over multiple years/sites and for two N inputs, prior to final mapping and definition of QTLs.

In the autumn of 2009, the Diversity trial was planted with 25 varieties. Two of the double haploid lines were replaced with two others, and 2 elite varieties were removed (Hurley and Monopol) and three new varieties introduced (Gallant, new high yielding and high protein variety; Oakley, highest yield RL Nabim group 4; Chablis, previously only assessed in 2004 and show anomalous responses at low versus high N inputs).

Objective 9: Drought Tolerance (UoN):

Background

Currently about 30% of UK wheat is grown on drought-prone land and drought losses are on average 1-2 t ha⁻¹. With climate change mean summer rainfall is expected to decrease in the UK by 20% by 2030, potentially increasing these losses. In addition to enhancing yield stability, improving water-use efficiency (WUE; above-ground biomass / crop evapotranspiration) will decrease crop water consumption in non-

drought years. This will increase water returned to the hydrological system for reuse, conserving water resources for use in irrigating other crops, and increasing water flows in rivers and water levels in wetland areas, as well as aquifer recharge. The activities under Objective 9 aim to identify key traits underlying improved water uptake, WUE and grain yield under drought. The underlying genetic basis of the key traits will be elucidated, because that information will greatly facilitate the deployment of the traits in improved germplasm. The identification of traits and markers to improve drought tolerance was not previously addressed in WGIN1. This research should over time greatly improve the resilience of the wheat crop to the anticipated effects of climate change and hence improve overall yield and quality stability.

Specific objectives

The specific objectives of Objective 9 are:

1. To identify the physiological traits explaining improved water-use efficiency and drought tolerance in elite winter wheat varieties.

2. To identify robust QTLs for water-use efficiency and drought-tolerance traits using one existing DH population in an elite background.

3. To develop one new DH population in an elite modern background segregating for drought-tolerance traits.

4. To identify novel genes and alleles controlling water-use efficiency and drought tolerance using the AE Watkins and Gediflux collections.

5. To collate a diverse germplasm collection (cultivars, advanced lines) from worldwide drought-tolerance wheat breeding programmes as a resource for future association genetics studies.

Summary of progress to date

The work in 2009 has focused on specific objective 1, according to the timetable for the milestones for the Activity 9. Previous work funded by DEFRA (projects CC0370 and LK0986) has identified varieties with contrasting yield responses to drought stress and WUE in field trials (irrigated vs unirrigated conditions). Thus, 18 varieties were chosen in consultation with breeders to represent contrasting drought performance mainly according to results from DEFRA project LK0986. The selection of the 18 varieties also included eight common varieties with the NUE variety phenotyping trial at Rothamsted in 2009/10 (see Objective 8). The work under specific objective 1 will quantify the size of varietal effects on water use, WUE and dry matter growth through the growing season and identify physiological traits underlying genetic variation in yield responses and WUE under drought. The key candidate target traits include: 1) deeper rooting, 2) high accumulation and remobilisation of stem soluble carbohydrate reserves, 3) optimisation of stomatal aperture traits (including stomatal conductance) underlying water-use efficiency and 4) delayed senescence with the stay-green trait.

A winter wheat field trial was successfully established at University of Nottingham in autumn 2009 on a sandy loam site to investigate associations between physiological traits and performance under drought for 18 elite winter wheat varieties at two irrigation treatments. The field experiment uses a split plot design with irrigation treatments randomised on main plots and 18 varieties randomised in sub-plots. There are three replicates, with a sub-plot size of $1.6 \times 12 \text{ m}$.

Irrigation treatments:

1. Fully irrigated (trickle irrigation system)

2. Unirrigated

Variety treatments:

1. Avalon	10. M. Widgeon
2. Beaver	11. Oakley
3. Cadenza	12. Panorama
4. Capelle-Desprez	13. Paragon
5. Cordiale	14. Rialto
6. Glasgow	15. Savannah
7. Hereward	16. Soissons
8. Hobbit	17. Xi19
9. Istabraq	18. Zebedee

An experimental protocol has been written for the scheduled assessments in 2009/10 including the following measurements in all sub-plots:

- Combine grain yield, yield components
- Above-ground biomass and partitioning at GS31, GS61, harvest
- % stem water soluble carbohydrate at GS61+10d
- Leaf senescence kinetics for flag-leaf, L2 and L3
- Water-use efficiency estimated by 13C isotope discrimination of grain dry matter samples

Further measurements will be carried out for a sub-set of six varieties showing the most consistent contrasts for drought tolerance according to data from LINK project LK0986.

- Stomatal conductance/photosynthetic rate using Licor 4600 gas exchange analyzer (subset of 6 varieties only)
- Crop water uptake from assessments of volumetric soil water content using the Delta-T PR2 capacitance probe (subset of six varieties only)

In addition, for the varieties that are common with the NUE phenotyping trial at Rothamsted N content of straw and grain at harvest will be assessed.

With regard to specific objective 9.4, an outline protocol for the assessment of visual traits (leaf green area and leaf rolling) on the AE Watkins lines in 2009/10 has been discussed and agreed with colleagues at JIC.

A summary of the research activities under Objective 9 was given at the WGIN management meeting at University of Nottingham on 20 October 2009.

Staffing

Mr Jayalath De Silva has been appointed at University Nottingham from 1 October 2010 (30 months) to work on the WGIN2 drought tolerance sub-project.

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

10.1 To screen the AE Watkins Collection and an 'improved' Gediflux Collection to identify potential sources of resistance to the take-all fungus under field conditions.

In the 1930's, A.E.Watkins collected around 4,500 wheat lines from around the world, which gave a snap shot of the germplasm available at that time. Over time the collection deteriorated, but at JIC they have rejuvenate approximately 800 lines. This unique collection may contain alleles and allelic combinations for disease resistance that breeders may have left behind. To explore this possibility, in the autumn of 2007 and 2008, 740 of these lines with sufficient seed were sown in the field in a high take-all risk situation at RRes. Due to shortage of seed, only 3 x 50cm length of row was possible for each line and these together with 5 blocks of eight controls (containing 5 current grown winter wheat, winter rye, triticale and oats) plus a further 20 plots of a fully susceptible wheat were arranged in an alpha designed experiment, total 800 plots (WGIN newsletter November 2008). Only herbicides and growth regulators were used to control weeds and crop height, respectively, no other pesticides were applied.

Weather conditions in 2008 were favourable for leaf diseases and advantage taken of this to score all plots for mildew, septoria, brown and yellow rust to assess for field resistance to these diseases. Differences between the lines did occur and results of these assessments can be found on the WGIN website. Plant samples were taken in July 2008 for stem base (eyespot, sharp eyespot and brown foot rot) and root disease (take-all) assessments. In 2009 weather conditions were not favourable for foliar disease development (see below).

In the autumn of 2008 sowing also included were 60 lines from the Gediflix collection (a newer collection, dating back to the 1950's). In addition to the herbicides and growth regulators, an eyespot specific fungicide was applied at T1, no other fungicides were applied. Although septoria and mildew were present on the lower leaves in March 2009, these diseases did not develop significantly due to unfavourable weather conditions until late in the growing season. However, in July all plots where leaf diseases occurred were recorded. Plant samples were taken at the beginning of July for take-all root assessments.

All stem base and root disease assessments from the 2007/8 experiment have been completed and are currently undergoing statistical analysis. Initial findings show over 200 lines were fully susceptible to take-all and these were discarded from the 2009/2010 autumn sown experiment. The remaining Watkins lines plus the new others from JIC which have not been previously tested and the 60 Gediflux lines were sown in another alpha design experiment as per previous years in a high risk Take-all field at RRes.

10.6 Generation and screening of mapping populations

Two of the *T. monococcum* populations have been taken to the F3 generation and seed is now available for screening for ~85 lines per population. All these lines are now being advanced to F6 by SSD. In total 6 Tm accessions have been identified which show the highest level of resistance to take-all. A detailed reciprocal crossing programme is in progress with these lines to a number of key susceptible genotypes. DArT marker analysis is partially complete and the subsequent PCA has reveals some of the resistant accessions are genetically quite close to fully susceptible accessions whilst the others resistant accessions are genetically unrelated.

10.7 Introgression of non-hexaploid and *T. monococcum* sources of resistance into hexaploid wheat

T. aestivum x *T. monococcum* crosses were completed using Avalon and Cadenza with embryo taking place at 14 days post pollination. In total 28 F1 rooted plants were transferred to the glasshouse after 4 weeks in vernalisation.

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

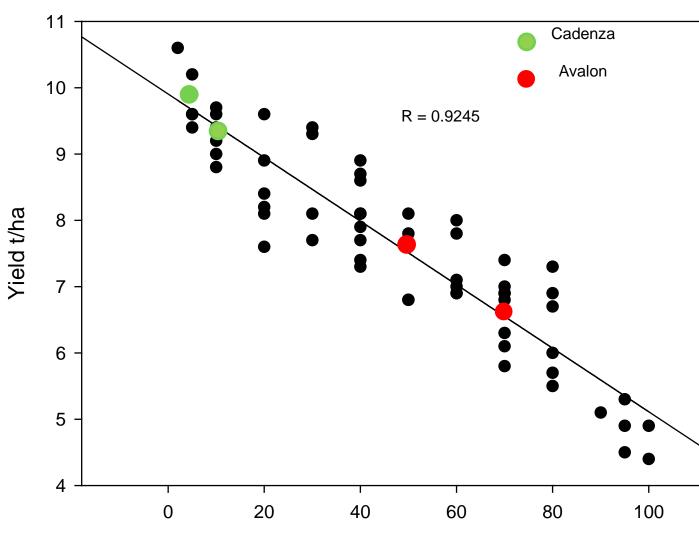
The diversity trial which is designed to examine nitrogen use efficiency of different bread wheats (see objective 08) contains 24 different wheat varieties grown as a first cereal crop. Advantage has been taken of this trial to explore the take-all inoculum build up in the soil under these different varieties. Soil cores were taken after harvest from all the different variety plots receiving 200k/ha of nitrogen. These cores were used in a bioassay using a fully susceptible wheat variety (Hereward) as the bait plant. Assessment of take-all has been completed and data is currently undergoing statistical analysis.

10.9 Explore the genetic basis of take-all inoculum build up using the Avalon x Cadenza mapping population.

Results from WGIN 1 have shown that Cadenza consistently builds up less take-all inoculum in the soil than Avalon when grown in a first wheat situation. The Avalon x Cadenza mapping population experiment (see objective 10.8) provides an opportunity to explore the possible genetic basis of take-all inoculum build up. Soil cores were taken after harvest from all 204 lines plus the parental controls. These cores were used in a bioassay using a fully susceptible wheat variety (Hereward) as the bait plant. Assessment of take-all has been completed and data is currently being used in a QTL analysis (see below).

The multiplication of seed for the Avalon x Cadenza mapping population is done on a separate field to the experiment and is grown as a first wheat crop. In one of these fields, the site was over drilled with the cultivar Oakley and data on take-all and yield has been obtained from the 67 different double haploid (DH) lines from the Avalon x Cadenza mapping population and 4 control parental plots grown the previous year. A strong correlation was identified between the final take-all disease levels in the Oakley crop grown over each of the original A x C DH plots and the final yield of these plots (**Figure 1**). There was ~2.5 tonnes advantage evident in both the Cadenza parental plots and many of the DH lines compared to the Avalon parental plots and many of the other DH plots.

Avalon x Cadenza lines in 2008, oversown with wheat cv. Oakley in 2009. Relationship between take-all patch score and yield



Take-all patch score (% area of plot affected)

Figure 1. Take-all inoculum build up trial 2008 - 2009 at RRes. Plotted is the take-all levels observed in the 2^{nd} wheat situation against yield.

Taking advantage of the extensive molecular marker data which has already been generated at JIC with the WGIN project for the A x C mapping population (see objective 03 (above), we have already been able to identify two major QTLs that confer both the take-all reduction and yield increase (**Figure 2**).

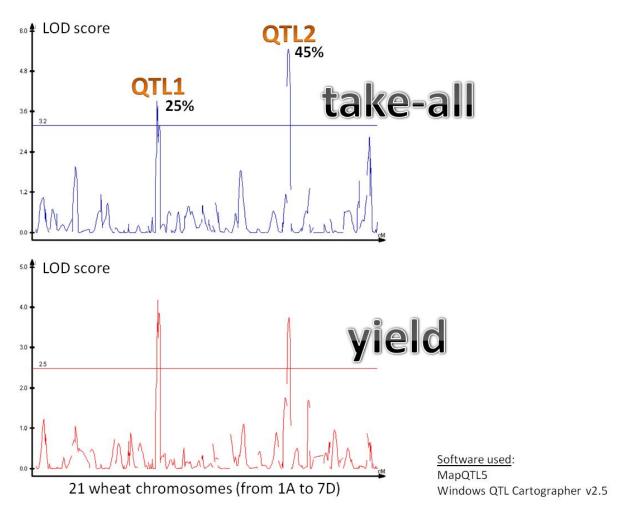


Fig.2 QTL analysis of take-all and yield

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

11.1 Convert the two closest linked DArT markers to the *TmStb1* locus to PCR based markers. It is not possible to obtain the DArT marker sequences from Triticarte, despite 6 months of repeated trying. Therefore we have switched to using the two closest SSR markers at 0.4cM and 1 cM.. These markers are robust and can now be compared to sequences available from the Brachypodium sequencing project in this genetic interval.

11.3 to 11.8 All genotyping and phenotyping were delayed due to a change in personnel in June 2009 and the replacement not arriving at Rothamsted till Nov 2009. Dr Hai-Chun jing who was an intergral part of the WGIN project since March 2004 left Rothamsted in July to become a programme leader at the Centre for Bioenergy Plants Research and Development, Institute of Botany, Chinese Academy of Sciences, 20 Nanxincun, Xiangshan, Beijing 100093, to research abiotic stress in sorghum. Temporary hire Steve Freeman and new permanent hire Dr Wing Sham Lee trained at Cambridge University will now be trained and collaborate with Kostya Kanyuka and Kim Hammond-Kosack to achieve these objectives. We have already

identified that only the external swab inoculation method is robust in the identification of the resistance phenotype.

Objective 12. Interconnections between the three soil-based explored traits (RRes and UoN) This final research object will not formally start until project year 2.5, but we have ensured that in the year 1 and year 2 NUE, WUE and take-all trails that there are common genotypes for cross referencing purposes in preparation for the combined data analyses.