WGIN 3 Management Meeting 1
4th March 2015 @ Rothamsted Research, Harpenden

This was the first Management Meeting of the newly **DEFRA** funded **WGIN3**.

Final Minutes

Attendees:-
Peter Shewry (PS) (chair), Malcolm Hawkesford* (MH), Andrew Riche (AR), Lesley Smart* (LS), Kostya Kanyuka* (KK), Vanessa McMillan* (VM), Kim Hammond-Kosack* (KHK), Michael Hammond-Kosack (MHK) (**RRes**), Simon Griffiths* (SG), Claire Lister (CL), Alba Farre Martinez (AFM) (**JIC**), Ed Flatman (EF) (**Limagrain**), Jacob Lage (JL) (**KWS**), Sarah Holdgate (SH) (**NIAB**), David Feuerhelm (DF) (**Syngenta**), Stephen Smith (SS) (**Elsoms**), Paul Meakin (PM) (**KTN**), Matt Kerton (MK) (**DSV**), Richard Jennaway (RJ) (**SaatenUnion**), Ruth Bryant (RB) (**RAGT**), David Cooper* (DC)(**Defra**) [*=giving presentation]

Apologies: Jayne Brookman (KTN), Dhan Bhandari, Ellie Marshall (HGCA), Simon Penson (Campden BRI), Simon Berry (Limagrain), Giulia Cuccato (Defra)

A. Welcome – Peter Shewry

B. Presentations:

1. Introduction to the new WGIN 3 project (KHK)

Prior to the application and funding of WGIN 3, two key documents were compiled and submitted to DEFRA. The WGIN legacy document covering the whole 10 years of WGIN 1 & 2 (in June 2014) [nb: to be put on the WGIN website by MHK shortly] and the final report on the 5 year WGIN 2 (in November 2014). The application for WGIN 3 was submitted to DEFRA in December 2014.

The key emphasis of WGIN 3 is **yield** stability and the working title of WGIN3 is: *Defra Wheat Genetic Improvement Network - Improving the resilience of the wheat crop through genetics and targeted traits analysis*

There are **four work packages** (details on presentation slide), **WP1** – Management, Networking & Communication, **WP2 & 4** – Genetic and QTL analyses of targeted traits and **WP3** – Tools and Resources. WGIN 3 consists of **2 project partners**, The John Innes Institute and Rothamsted Research and **3 new subcontractors**, Bristol Genomics Facility (University of Bristol), Affymetrix and MYcroarray (Michigan, USA).

WGIN 3 comprises **21 milestones**, with individual target dates between March 2015 and December 2016 (details on presentation slide).

2. Genetic Resource Development for UK wheat yield stability (SG)

Consistency of high mean yield levels of wheat and identifying the genetic traits that deliver this are increasingly important targets. To dissect the genetic gain of wheat the following

questions need to be answered: What genes control these traits? How do alleles work in combination for genetic gain and trait stability? The key platform to start answering these questions are Near Isogenic Lines (NILs), many of which have already been generated during WGIN 1 & 2. To achieve high mean yield it is very important to understand the basis of QTL x environment interactions. SG's group are going to look at historical climatic data regarding the environmental impact on yield with the aim of establishing gene hypotheses to put into the breeding pipeline. Indeed, WGIN has already validated a lot of QTLs.

Experiments assigned to WGIN 3 are
(1) A chromosome segment substitution library for Avalon x Cadenza (AxC) (**WP3.2**)
(2) Applying WGIN data to breeding by design for UK yield stability (**WP3.2**)
(3) Dissecting UK drought tolerance in Paragon x Garcia (**WP2.3, WP4.3, Milestone 19**)
(4) Quantifying agronomic impact of WGIN target genes using the Paragon NIL library (**WP2.3, WP3.2**)
(5) Informing multiple marker assisted selection for yield stability using Paragon library (**WP3.2**)
(6) Foundations for a new generation segregating populations for studying yield stability in the UK (**WP2.1**)

C. DC stressed the need to emphasise how WGIN 3 and yield stability affect UK competitiveness and economic growth and to communicate this to as wide an audience as possible. Workshops and stakeholder meetings are very important for this. Politicians and the general public need to be made aware of this. [nb: these comments apply to all of WGIN, not just SG's part]

Q. DF – are all field trials conducted under full fungicide applications?
A. yes
C. DF – this needs to be emphasised, because the greatest effect on yield stability would be fungicide withdrawal.

C. JL – very good that WGIN is looking at historical data. The more of this the better, because there is a strong need to get a handle on why QTLs occur then disappear.

Q. PM – with climate change (wetter winters, drier summers), why is WGIN concentrating on drought tolerance only?
A. SG – not just pinning drought down.

3. Stability/Resilience (MH)

MH will continue the Nitrogen Use Efficiency (NUE) trial during WGIN 3. More than 50 wheat varieties at four nitrogen levels have been grown for 11 years already and will be grown for the next few years (25 per year with a core of 10 varieties grown every year), but not all data have been exploited yet. The analyses will reference climatic data and also include long term data from Broadbalk.

Post anthesis mineral uptake (N but other minerals as well) is also being investigated to establish a link between mineral uptake and grain protein deviation (GPD).

MH and AR are continuing to use their UAV (unmanned aerial vehicle) to assess canopy longevity (using the green normalised difference vegetation index, GNDVI) and to measure plant heights. While there is a very good (R^2=0.99) correlation between measurements of plant height with tape

measuring, the UAV advantage is speed (10min programmed flying time, 1-2days CPU for 3D assembly but no person-hours involved) and accuracy.

Q. JL – are all these data in an accessible format on the WGIN website?
A. MH – no, because still not entirely happy with the data, BUT available on request
C. KHK – MTA for AxC population could be used as a blueprint for an MTA to access specific unpublished sets from the Diversity trial
Q. MH – did anyone present want any specific wheat line tested?
A. no one came forward
C. KHK, MH – over 8000 grain samples (500g each dried to 12% moisture, stored at -20^{o}C) from these trials available upon request

4. Screening germplasm for resilience to aphids (WP2.3) (LS)

Two species, the bird cherry-oat aphid *Rhopalosiphum padi* and the grain aphid *Sitobion avenae* have been used for these studies. Previous experiments have been carried out within the BBSRC funded LOLA and WISP projects. Diploid wheat lines have been far more effective than hexaploid ones in reducing nymph numbers and development. *Triticum monococcum* lines show the best resilience. Over 40 *T. monococcum* lines have been assessed for aphid resilience, and two lines, MDR045 and MDR657 had not a single nymph (for both species) whereas MDR037 showed high numbers and good development for both species. MDR049 takes longer to accumulate nymphs. These lines have also been assessed in electrical penetration assays (EPG, details in online presentation) where MDR049 and MDR657 where shown to increase the E1 wave form significantly. This wave form is associated with salivation prior to ingestion (by the aphid, not the researcher). Wave form E2, which is characteristic of phloem ingestion, was reduced on these lines indicating interrupted feeding consistent with the observed reduced development.

Work in WGIN 3 will involve phenotyping of F_1 plants of *T. monococcum* crosses already generated by MHK (MDR037(susceptible) x MDR045(resilient), MDR037(susceptible) x MDR657(resilient) and MDR037(susceptible) x MDR049 (semi-resilient)). Further generations (F_2 and beyond) will be generated by MHK and phenotyped by LS.

Q. DC – is there a difference between virus carrying aphids and non-carriers.
A. LS – could be tested with antibodies or PCR

C. KHK – *T. monococcum* lines, MDR037,MDR045 and MDR049 originate from Vavilov Institute, Russia (described in WGIN publication, Jing et al., (2007) *Journal of Experimental Botany* 58, 3749-3764.

Q. KK – is there a link between leaf morphology (a lot of *Tm* lines have very hairy leaves) and resilience?
A. LS – nothing obvious, nymphs can easily move between leaf hairs.

C. LS – emphasised that diploid wheat species are far more resilient than hexaploids.

Q. JL – is there any prospect of actually developing resistant commercial wheat lines? Should the focus be on hexaploid lines.
A. LS – not sure, this research is far behind other traits, ie there are no QTLs available.

C. just in case anyone was wondering – it was mentioned that aphids can form part of a healthy human diet and taste sweet…

5. Resistance to take-all and foliar diseases (VM)

For WGIN 3 there are four objectives including
[1] complete development of *Triticum monococcum* mapping populations for genetic analysis of root resistance to take-all: Two populations are already at the F_6 stage and two more will be taken from F_4 to F_6 (details in online presentation)
[2] continue the introgression of resistance to take-all from *T. monococcum* to the BC1 stage (done by MHK): Five *Tm* lines were crossed to Paragon *ph-1* mt resulting in a total of 290 grains. In total 30 grains have been embryo rescued (none germinated) and so far one F_1 plant has developed but is infertile (both male & female, although both anthers and stigma organs were fully formed)
[3] examine the resistance of *Triticum monococcum* to yellow rust: because although *T. monococcum* has been grown at RRes since the start of WGIN 1 in 2004, no obvious yellow rust infections were observed. The total *T. monococcum* collection (263 accessions) was sown in a field trial last autumn and will be scored for yellow rust during WGIN 3. Also, the entire *T. monococcum* collection will be genotyped by Bristol Genomics Facility.
[4] characterise hexaploid wheat germplasm previously shown to exhibit a high level of resistance to multiple foliar diseases: during WGIN 2 a 3rd wheat, Take-all field experiment in 2008 (Richard Gutteridge) using the Watkins collection (740 lines) was conducted without applying any fungicides and assessed for yellow rust, brown rust, septoria and powdery mildew infection and plant samples taken for take-all assessments on the root systems. Ten Watkins lines were resistant to all four foliar pathogens. These lines have been sown in both 1st wheat (no take-all) and 3rd wheat (high take-all) field trials in autumn 2014 and will be scored for all four foliar diseases as well as Take-All, to establish whether foliar disease resistance is triggered by plant defence responses against Take-All.

C. DF – useful to compare historical data to 2008 trial
C. SG – there are some very old yellow rust data. Also, a Sydney group has used the Watkins collection to score for yellow rust resistance.

Q. JL – Where are are all 10 Watkins lines originating from?
A. All are from SG (JIC). The line numbers are 18, 137, 203, 231, 262, 399, 495, 610, 733 and 786.

C. DF – transfer from *Tm* to hexaploid likely to lead to just another "boom and bust" cycle

C. JL – *Tm* introgression should be dropped, because everyone has tried it and it has never worked, but if you were to establish a technique this would be a major breakthrough.

6. Septoria (*Zymoseptoria tritici*): Resistance from *T.monococcum* (KK)

In total, 18 major genes (Stb) for resistance to *Z. tritici* and 36 resistance QTLs have been identified in wheat. During WGIN1, 120 *T.monococcum* lines were screened for resistance to 9 Septoria isolates. MDR308 was shown to be resistant and MDR002 highly sensitive. A cross between MDR308(R) x MDR002(s) showed that F_1 plants were resistant to isolate Zt IPO323. 94 F_3 families were screened for segregation of resistance / susceptibility to Zt IPO323 and it was shown that resistance to *Z. tritici* IPO323 in *T. monococcum* MDR308 appears to be inherited by a

single gene, *TmStb1*, which was mapped to the top arm of chromosome $7A^m$. Because of difficulties encountered during phenotyping, these F_3 lines will be re-phenotyped during WGIN3. The reverse cross was also carried out and phenotyping of this *T. monococcum* MDR002(s) x MDR308(R) population has been performed. 79 F_3 families have been screened and 30 F_3 were fully resistant, 25 F_3 fully susceptible and 19 F_3s segregated for resistance.

Q. DF – are there any plans to transfer this resistance to hexaploid wheat?
A. KHK – yes, this is going on already (by MHK)

7. Exome capture by MYcroArray (KHK)

Exome capture forms part of **WP 4.2, 4.4** and **Milestones 18**. Just in case anyone was wondering, "an exome is to a genome as an abstract is to a research article: concise, information-rich, and easily digested" (JM Perkel), being the protein coding content of, and only comprising 1-2% of the genome. The sub-contractor selected for this is the company MYcroArray, based in Ann Arbor, Michigan, USA. This company uses MYbaits, which is a fully customisable liquid-phase DNA capture system for targeted sequencing, and liquid-phase capture is apparently more efficient.

The overall aim during WGIN3 is to employ exome capture to identify genetic variation in candidate or known genes that are responsible for the desired trait (s). A designated group of WGIN scientists will interact with the company MYcroarray to decide on the best way to represent wheat genes on the 20,000 bit array. This will be done via a series of Skype meetings held during months 1-3.

Importantly, a workshop will be held to prioritise the gene list and the 96 wheat genotypes to be tested. The priority gene list (and corresponding oligo design) will involve discussions within WGIN. The first year (2015) will include all discussions, workshops and manufacture of the oligos. The actual exome capture will be carried out in year 2 (April 2016) at MYcroarray. Also, it is intended to interact with the BBSRC funded BBR project which already includes some exome capture for wheat (Uauy (JIC) and Philips (RRes)).

C. JL – the 96 wheat genotypes selection should be a WGIN community wide activity and involve a concrete plan for subsequent data sharing.

A1. KHK – all data could/will be hosted at Bristol Genomics Facility
C. JL – great idea
A2. A small group of 3 WGIN scientists and 3 WGIN breeders will select the 96 wheat varieties.
C. DF – the key to success is good communication and easy data access.

C. The oligo design will be carried out by a small group of WGIN scientists, and also involve Cristobal Uauy (JIC)

C. SG – very keen to be part of this phase
C. SH - nominated Alison Bentley at NIAB to be involved

8. Defra and the new industrial led forum (DC)

Government ministers change priorities according to political climate, but the overriding factor regarding funding for science is "increasing economic growth". WGIN needs to find a balance between what ministers want to see and the interesting science. WGIN needs to become more visible and be seen as involving (instead of excluding) Agritech industries. [SG is about to meet with ministers to increase awareness of the existence of WGIN]. However, there is a Catch 22 situation – if WGIN turned out to be commercially viable, then according to ministers, industry should take over and government funding would be withheld. Safeguarding plant health, resilience (to climate change) and the sustainable use of indigenous resources are currently funding priorities.

WGIN does have an important part to play prior to Agritech and is a necessary part of the *innovation pipeline*. The annual "Cereals" exhibition is a good opportunity to highlight this and make farmers and the wider wheat community aware of involve them more in WGIN.

C. DF - Peter Gregory (HGCA) who chairs the cereal evaluation committee should be invited to WGIN meetings.

DC stressed repeatedly that the WGIN's public visibility is not high enough. For example, the "Plant Breeding Matters" document (BSPB) (circulating in parliament) does not mention WGIN. Giulia Cuccato (Defra) is currently putting together a document to highlight the importance and influence of WGIN. Generally, the audience outside the WGIN community needs a "cartoon approach" similar to KHK's cartoon explaining exome capture.

C. PS – use the RRes communications department to publicise WGIN

C. already need to start thinking about another extension of WGIN at the end of this year.

C. Stakeholder Meeting

The next WGIN stakeholders meeting will take place on **Thursday April 16th 2015**. The venue will be JIC (the meeting room in The Genome Centre (capacity ~65)). The presentations should emphasise the achievements during WGIN 1 & 2.

C. MHK has sent an initial email on March 4th (pm) to stakeholders, breeders and scientists with the date for the stakeholders meeting.

This date will also be used for the first small group workshop to determine the 96 wheat varieties for the exome capture (see B 7(KHK)). The stakeholder meeting will take place in the morning and finish with lunch, and the workshop will take place in the afternoon.

<u>Action:</u> Two more scientists (SG already selected) and 3 breeders need to be chosen for the workshop asap

It was agreed that because of time limitations a big push to increase WGIN's visibility should be made for the next stakeholders meeting in November to approach people directly, including journalists and institute directors.

D. Management Meeting

The next management meeting was agreed to take place around July 20th 2015. To determine everyone's availability and thus the exact date MHK has emailed a Doodle poll to everyone in the management meeting group on March 11th 2015. Date fixed for Friday 17th July 2015 @Rothamsted Research

E. Note - All the ppts from this management meeting have already been uploaded onto the WGIN website

F. A funded overseas workshop application to BBSRC

Peter Shewry to investigate the possibility of holding a joint wheat workshop in Russia in 2016. To include The Vavilov Institute, St Petersburg and possibly one other Institute.

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