WGIN 3 Management Meeting
20th April 2017 @ Rothamsted

This was the eighth Management Meeting of the **DEFRA** funded **WGIN3**.

Minutes

Attendees:
Peter Shewry (PS) (chair), Lesley Smart* (LS), Andrew Riche* (AR), Vanessa Mcmillan* (VM), Kim Hammond-Kosack*(KHK), Michael Hammond-Kosack* (MHK) (RRes), Simon Griffiths (SG), Clare Lister* (CL) (JIC), Dhan Bhandari (DB) (AHDB), Ed Flatman (EF) (Limagrain), Matt Kerton (MK) (DSV), Sarah Holdgate (SH) (NIAB), Lucy James (LJ) (ADAS), Martin Cannell (MC)(Defra)

[*=gave oral presentation]

Apologies:
Malcolm Hawkesford (RRes) Jacob Lage (KWS), Ruth Bryant (RAGT), Stephen Smith (Elsoms), Simon Berry (Limagrain)

A. Welcome – Peter Shewry

B. Presentations:

1. Review of minutes from January 16th 2017 (KHK)

Approved by all. To add to website.

ACTION ITEM – KHK to interact with Paul Kersey at EBI, to place the new TILING data from Clare Lister on the ENSEMBL wheat genome as a separate track

2. Tools, resources, genotyping and phenotyping – (CL)

Details can be found in the presentation (pp1-28) on the WGIN website.

The Garcia – Paragon drought trial

In the 2015-2016 season explore senescence using Drones. The images have been analysed by PI Chris Applegate (Earlham) focussing on excess green minus excess red. Focused on the central 3 rows of each plot, taken from a close up image. 15 of 18 plot times have been explored. Waxy vis non-waxy lines were very clearly visible.

The QTL analysis of the data, indicates that from 10th June onwards, the same QTL (for what add) appears earlier in the non-irrigated plots. Samples have been taken from this trial for metabolite and transcriptome analysis to determine what features are linked to senescence. This additional study is supported by another funding source.

CIMMYT manual –
http://repository.cimmyt.org:8080/xmlui/bitstream/handle/10883/1287/96140.pdf
Physiological Breeding I: Interdisciplinary Approaches to Improve Crop Adaptation
http://repository.cimmyt.org/xmlui/bitstream/handle/10883/1288/96144.pdf
Physiological Breeding II: A Field Guide to Wheat Phenotyping.

GS30-GS31 and the start of elongation, is when grain number is being determined. GS31 to booting is the most important for drought accelerated senescence.

The 2016/2017 Garcia x Paragon trial has been drilled on a lighter sandier soil type
Already 4 lots of watering have taken place using a spray boom. In April a lot of the plants only had 20 cm of roots

Q EF - Are you exploring in this trial drought tolerance or drought avoidance?
A-

Q MG – You have identified major QTLs, how will these be taken forward
A- Will work on the larger blocks and break these up into sub-lines to explore further through field experimentation

New field technology
Clare demonstrated during the coffee break a new technology for the field that had ben identified during a recent visit to CIMMYT. The |KDSmart Field Scoring App and can be downloaded from Googleplay for free.
"KDSmart" is a very good field–scoring app from "Diversity Array Technology", designed for use on an Android device. It is used at and recommended by CIMMYT. A field plan, information about the trial and traits to be scored are loaded onto the device and data can be directly entered while in the field, and then uploaded in the lab.
It is recommended to download the demo trials and become familiar with the app in plenty of time before needing to use it!
www.diversityarrays.com/kddart and
www.kddart.org/help/kdsmart/

CSSL library – Avalon x Cadenza

Ongoing. 20 Mb a representative genotype – the grid result based on NR values.

3. Using the WGIN Diversity trial to develop drone applications (AR)

Details can be found in the presentation (pp29-39) on the WGIN website.

Initial focus of the presentation was on disease detection not using the diversity trial

Explored different altitudes. At low heights the plants moved , there a longer lens could be tried to obtain the data
Camera in use with a 24 MPix APSC –sensor

Overall conclusion – please add

Diversity Trial - 30 cultivars just group 1 and group 4 wheats
Soweing date – add into minutes

C PS You also need to plot the nitrogen recovery in the grain for each cultivar

C PS a 10 tonne crop will need ~250 kg/N applied

Q DB Will you be focussing on any other NABIM group wheats. No just groups 1 and 4.
ACTION Item ALL Agreed discussion topic for the next Management meeting. Should the N regime for the diversity trial be changed starting in autumn 2017? The rates to be considered are N0, N150, N200, N250, N300, N350.

C- Potassium is washed out from the grain in August if it rains and the crop is left to stand.

C- KHK Hybrids what is the effect on the various traits explored, could these cultivars be highlighted in the graphs.

4. Resistance to Aphids (LS)

Details in Presentation online (pp40-48).

Overall the better phenotyping data has been achieved using the aphid *S. avenae*. The *R. padi* results are always less consistent.

Now have Barley Yellow Dwarf virus (BYDV) free isolates for both species

Commercially, *R. padi* may give a greater yield losses because of the virus transmission.

S. avenae tends to go into the ears at anthesis

C-VM – field grown *T monococcum* lines have not been infected with aphids so far in 2017

Q- Seedling tests, death or just arrested growth/ development
A- With line MDR 49 the nymphs are still alive and the next generation does eventually develop.

5. Development and exploitation of *Triticum monococcum* germplasm resources (VM)

Details in presentation online (pp49-76)

In the 2016-2017 field season, the lines most highly resistant to yellow rust are so far susceptible to brown rust.

Line W610 will need to be re-scored at a later date than the others because of late flag leaf emergence.

Lines W203 and W610 are showing good resistance to all three diseases (yellow and brown rust plus Septoria)

Line W786 has a high number of tillers. It exhibits adult plant resistance, the same as in the previous season. Because of this the crosses are 1 year behind.

Overall disease situation 2017 At Rothamsted the south site of the experimental farm has good disease, but the north side has very little disease. At RRes there was a lot of early yellow rust disease which then went away. Currently yellow rust, a lot of brown rust and low to moderate septoria. No mildew

Q- Do you plan to screen for the presence / absence of Yr15.
A. Yes

6. Update on Wheat Promotome Capture (MHK)

Due to an overall reluctant to omit any of the genes submitted by the trait coordinators (31 genes too many for a MyBaits1 custom set), a closer look at the costs revealed that it was possible to go for a MyBaits2 set with 40,000 MyBaits probes. Thus **all** gathered genes, plus a few additional ones provided by Sigrid Heuer, could be included, as well as extending the promoter sequence lengths to **1700bp** (previously 1000bp).

The final version of the WGIN Promotome FASTA has just been completed, using the 'gold standard' **Wheat IWGSC RefSeq v1.0 chromosomes** as a reference. All 22 chromosomes (Chrs 1-7 ABD plus ChrU (still unassigned sequences)) were downloaded from the URGI website (http://urgi.versailles.inra.fr) into the Geneious software. CDS, starting from the ATG start codon, were obtained for all genes from their Ensembl gene IDs using Ensembl Biomart (http://plants.ensembl.org).

Generally, the first 70bp of each CDS was used to search the respective chromosome with 100% homology. 1700bp of promoter/utr sequences were selected and copied into an Excel spreadsheet, noting the physical location on the chromosome and the orientation (forward or reverse). If the gene was not found, the search was repeated with the first 35bp. Any genes not found with the shortened sequence were listed as 'not found' and subsequently subjected to an URGI BLAST against **all chromosomes** (as opposed to just the chromosome specified in the ENSEMBL ID).

A summary of the data obtained is shown below:

1) A total of 1480 gene IDs were searched.
2) 1251 gene locations were found with 70mer CDS sequence
3) 105 gene locations were found with 30-35mer CDS sequence
4) 124 genes were 'Not Found'
5) Of these later 124, 89 were found in the URGI BLAST on **different chromosomes**. This included **all** IDs previously unassigned in Ensembl/TGACv1. In several cases the BLAST found homologues with 100% homology. In these cases the search was repeated using the complete CDS (varying from approx.1000 to 6000bp). In all these BLAST searches only 1 location remained with 100% homology and full length sequences.

6) The remaining **35 promoters**, not found in the **Wheat IWGSC RefSeq v1.0 chromosomes**, were therefore obtained from the Ensembl/TGACv1 database (release 34, January 2017). These 35 promoter IDs were labelled to include TGAC to make clear that the reference is **not** RefSeqv.1.0

In total, 1445 (97%) genes and promoters could be physically located on individual chromosomes.

Furthermore, the gene IDs were changed to include a reference to a) the chromosome location, b) a seven digit number from the original Ensembl IDs and c) the trait (T1 to T10) and gene number for each trait to facilitate referencing each sequence. To give just two examples (more details to follow elsewhere later):
1AL-0025400-T8-22 is gene 22 from trait 8 (Flower Biology), located on chromosome 1A and the original Ensemble ID TRIAE_CS42_1AL_TGACv1_001119_AA**0025400**.2

1AL-TGACv1_0037970-T7-20 is gene 20 from trait 7 (Canopy Development) **BUT** only found in the Ensembl/TGAC database and **not** the RefSeqv1.0 database.

Seed of all the 92/96 cultivars / genotypes is now at Rothamsted from various sources. DNA for the remaining four varieties was provided by RAGT. All the plants selected for tissue sampling and DNA extract have gone into vernalisation and seed will be collected only from bagged ears.

C- Need to check that the DNA extraction kit selected is fully compatible with the Mybaits technology.

Q- Who does the repeat masking
A- The company
C- Need to manually check some of the results post the repeat masker step.

C- If this approach is successful, in WGIN 4 could propose an intron scan. Thus WGIN would focus on the non-coding regions associated with the CDS.

ACTION Item all trait co-ordinators – to update the traits lists with publications

ACTION item : Next WGIN MM meeting to include a30 min slot in the agenda to discuss how to proceed with the sequencing data when it is returned by the company

7. Feedback on Stakeholder Meeting (PS)

The general view is that it was a good meeting and that the panel discussion was well received. But it was suggested to have slightly fewer presentations in future. And although the meeting was well attended, the stakeholder base is fairly static and it would be worthwhile to try to extend this. Towards this end it might be good to include a very short paragraph with each presentation title at the time the agenda was advertised. SH further

suggested to re-introduce the use of feedback forms to gauge the impressions of the stakeholders.

8. Date for the next Stakeholder meeting, 30th November 2017 at Rothamsted Research.

You can already register on line following the directions on the WGIN website.

In 2017 the event will be joint with the new BBSRC Institute strategic programme 'Designing Future Wheat' (DFW) to further enhance the synergies between the BBSRC and defra funded research.

C-MC - The GINs activities align very well to the BBSRC research

C-MC – also planning another joint GINs event. But not sure how much can fit into a single day. The aim will be to achieve a balance between the more technical talks and fulfilling the needs of users beyond the breeders. We would also like to involve more senior policy people from both Defra and the BBSRC to this event. AHDB will become involved in devising a joint GINs event in 2018. We would need to devise a draft agenda by mid-end Nov 2017.
Ideas - Could have just one large discussion with an invited panel, or split into smaller groups for a discussion/ consultation.

9. New funding, studentships using WGIN data and resources

The Excel spreadsheet will be circulated for updating in June 2017

10. New publications

None reported, several have been submitted and are under review.

11. AOB

Mike Ambrose the Germplasm Bank Manager at the JIC will retire in 2017 and a job advert for this full-time post is current out.