# Annual Project Report
# October 2020 to September 2021

| | | | |
|---|---|---|---|
| **Project title** | Developing systems to control male fertility in wheat for hybrid breeding, enhanced pollen production and increased yield | | |
| **Project number** | 21130024 | | |
| **Start date** | 01 October 2016 | **End date** | 30 April 2022 |

**Project aim and objectives**

In cereals, successful reproduction is critical to grain set and crop yield. Thus, optimising and enhancing fertility, alongside controlled fertilisation for breeding and hybrid development, is key to achieve high yields in a sustainable manner. As hybrid vigour increases yield, developing hybrid crops offers opportunities to increase productivity. However, this is challenging, due to the need to avoid self-fertilisation. Therefore, mechanisms that control fertility in a reversible manner are needed. There is also a requirement to ensure effective pollination, which relies on high levels of viable pollen for cross-pollination that is distributed effectively and is resilient to abiotic stress.

This project will provide a greater understanding of pollen development in cereals. It will develop switchable systems for the control of wheat fertility and identify traits for enhanced pollen production and viability (particularly under environmental stress), which are critical to ensure pollination in breeding programmes. The project will also determine the environmental stability of fertility-control mechanisms in barley and wheat to improve understanding of fertility control under various temperatures and light intensities. This work will help characterise the underlying fertility-control mechanisms. This is important due to the diploid nature of barley and the greater tools and resources available for barley genetic analysis. By investigating the mechanisms behind these traits and generating tools for breeding and selection, effective breeding to increase crop productivity and resilience will be realised. The work will also identify the benefit of hybrids for fertility control and the stability of these systems in elite breeding materials.

**Key messages emerging from the project**

There is conservation of the gene networks involved in pollen development. Genes have been identified from barley and wheat that are critical for pollen development. These are being targeted by mutagenesis to help characterise their function and to generate male sterile lines that could be used for hybrid breeding approaches. In addition, gene editing is being used to produce the same phenotypes as those generated by conventional mutagenesis. Mutants, where all copies of the genes have been mutated, are male sterile due to failure of pollen development. This phenotype is conserved for the mutants in barley and wheat lines.

# Annual Project Report
# October 2020 to September 2021

| Summary of results from the reporting year |
|---|
| 1. We have generated wheat triple TILLING mutants in which all copies of key transcription factors involved in pollen formation have been mutated. This involved the selection of mutant lines and crossing to combine the mutations over a number of generations until homozygous (Hm) mutations were obtained in the hexaploid bread wheat Cadenza. Genotyping and KASP-marker analysis was conducted throughout these crosses to ensure the backgrounds were correct and presence of the mutations maintained. We have done this in the Cadenza wheat background, in which the TILLING population was generated, alongside our Industry Partners, who have independently introgressed these mutations into their elite winter wheat breeding lines.<br>2. Preliminary analyses of Hm lines in the Cadenza background indicated male sterile phenotypes in the homozygous triple mutants. Further backcrosses have been conducted and homozygous stable mutant lines are being analysed to confirm the phenotypes by detailed microscopy and gene expression analysis. These wheat genes have been used in complementation analyses, using equivalent arabdiopsis mutants, to test whether the wheat genes are able to rescue the phenotypes of the corresponding arabidopsis mutants.<br>3. These mutants have also been transferred into three elite breadwheat varieties. Backcrossing of these lines is ongoing; triple mutants, carrying defects in each homeolog, are under investigation for fertility defects. Manuscripts on the wheat mutants study are in preparation.<br>4. In addition, we have generated the equivalent mutants for these genes in the diploid barley and have been characterising their phenotypes to determine the basis of the observed male sterility. |

| Key issues to be addressed in the next year |
|---|
| 1. Completion of phenotype analysis of Cadenza triple homeolog mutant wheat lines by microscopy and expression analysis.<br>2. Analysis of Cadenza triple mutants under various light regimes to test for induction/rescue of sterility.<br>3. Analysis of triple mutant wheat lines in elite backgrounds that have been generated by the plant breeding industry partners.<br>4. Completion of field screening and data analysis for male fertility traits. |

| | |
|---|---|
| **Lead partner** | Prof Zoe A Wilson; University of Nottingham |
| **Industry partners** | KWS, LIMAGRAIN, RAGT, SECOBRA |
| **Government sponsor** | BBSRC |

The results described in this summary report are interim and relate to one year. In all cases, the reports refer to projects that extend over a number of years.

# Annual Project Report
# October 2020 to September 2021

| Has your project featured in any of the following in the last year? | |
|---|---|
| ***Events*** | ***Press articles*** |
| | |
| ***Conference presentations, papers or posters*** | ***Scientific papers*** |
| | • José Fernández-Gómez, Behzad Talle, Zoe A Wilson (2020) Increased expression of the *MALE STERILITY1* transcription factor gene results in temperature-sensitive male sterility in barley. J Exp Bot. 71: 6328-39 DOI:10.1093/jxb/eraa382 |