



## Accepted Applications in the Community Resource for Wheat Transformation Project (2012-2017)

### **Calcium-mediated signal transduction; generation of calcium reporter lines**

Dr Julia Davies, University of Cambridge

Transient elevations of cytosolic free calcium in response to stimuli (e.g., stress, pathogens) are known from studies on Arabidopsis to result in specific transcriptional responses. In this project, wheat will be transformed to express (apo) aequorin in the cytosol as a reporter protein for free calcium to enable stimulus-specific calcium signals to be resolved.

### **Functional analysis of the cytoskeletal system in response to fungal pathogens**

Dr Mike Deeks & Prof. Patrick Hussey, University of Durham

Plants utilise the actin cytoskeleton to defend against biotrophic fungal pathogens. Compromising this cytoskeletal defence through drug treatments or through genetic manipulation of actin dynamics enhances the abilities of a broad range of pathogens to breach the plant cell wall. We have identified an actin binding protein from Arabidopsis thaliana that is an early marker of defence sites counteracting powdery mildews. We are constructing a fluorescent protein fusion to the wheat homologue of this protein and an RNAi knockdown line for the suppression of the endogenous wheat gene. This will allow us to monitor the response and defensive function of this protein to wheat-compatible infections.

### **Manipulating the epidermal patterning factor signalling pathway to reduce stomatal density and improve wheat water use efficiency**

Prof. Julie Gray and Lee Hunt, University of Sheffield

We have previously demonstrated that manipulating expression of Arabidopsis epidermal patterning factor genes can reduce stomatal development and enhance plant growth particularly under drought conditions. The aim of this project is to see whether manipulating similar genes in wheat could alter stomatal number, water use and yield.

### **Improving resistance to pre-harvest sprouting**

Prof. David Hanke, University of Cambridge

David Hanke's group has isolated a gene, StCKP1, from potato which, when expressed in Arabidopsis from either a constitutive or a seed storage protein promoter, prolongs the duration of seed dormancy. We will express the rice homologue of StCKP1 in wheat and characterise seed dormancy and resistance to pre-harvest sprouting.

### **Overexpression of the photoprotective gene PsbS in wheat**

Dr Erik Murchie, University of Nottingham

Photoprotection refers to a range of mechanisms that plants use to prevent damage when the amount of light absorbed by chlorophyll exceeds that required in photosynthesis. In fact, protein-chlorophyll complexes are capable of 'dissipating' excess energy but this process requires fine control to prevent a loss in photosynthetic efficiency. An important protein is called PsbS, which acts to regulate this process; in particular,



it accelerates dissipation in vivo. It has been shown that over-expressing PsbS in Arabidopsis is capable of preventing light-induced damage to leaves and in turn, determining productivity in the field. We intend to over-express the PsbS gene in wheat and test for improved stress tolerance and improved productivity under variable light conditions.

### **The role of the ubiquitin proteasome system in the interaction between wheat and the fungal pathogen *Mycosphaerella graminicola***

Dr Ari Sadanandom, University of Durham

The ubiquitin proteasome system (UPS) is the central modifier in all plant signalling and, as such, plays an important role in defence against pathogens. This project aims to investigate the affects of certain UPS components in the interaction between wheat and one of the most economically important wheat pathogens worldwide, *Mycosphaerella graminicola*.

### **Developing a cereal fertility pipeline (CerFip) for wheat and barley**

Prof. Zoe Wilson, University of Nottingham

Key advances have been made in understanding pollen biology using the molecular genetic resources available for Arabidopsis to dissect the pollen regulatory gene network. Transferring our knowledge of reproduction gene networks from Arabidopsis and Rice into barley and wheat will have future application in developing germplasm for selective breeding, for the generation of hybrids and therefore for increasing yield.

### **Manipulation of meiotic cross over frequency in wheat using FANCM-specific TALENs**

Prof. Keith Edwards, Cereal Genomics Group, University of Bristol

Site directed mutation of the Arabidopsis Fanconi Anemia, Complementation Group M (FANCM) gene, leads to an increase in the frequency of meiosis specific crossovers, an effect which is thought to arise by a reduction in cross over interference. Wheat varieties with an increased rate of recombination would have great potential

as breeding material. To modify the wheat FANCM ortholog, we will use TALEN-based technology to engineer wheat as previous described for Arabidopsis. The resulting lines will be characterised for a range of mutations and their effect on the rate of recombination will be monitored by the generation of specific mapping populations.

### **Assessing the potential of AVP1 to increase abiotic stress tolerance in wheat**

Dr Heather Knight & Prof. Marc Knight, University of Durham

The vacuolar type I proton pyrophosphatase AVP1 regulates ion and sugar accumulation in the vacuole leading to an increased ability to retain water. This has been associated with the ability to tolerate drought and salinity stresses. This protein represents an excellent candidate for the development of wheat strains with increased tolerance of abiotic stress.

### **Functional analysis of TaRSR1 in wheat grain development and starch metabolism**

Dr Sinead Drea, University of Leicester

We propose to functionally characterise the AP2-related RSR1 gene which has been shown to affect starch metabolism in rice where it represses expression of starch synthesis genes in the grain (Fu and Xue 2011 Plant Physiol 154,927). As well as examining the effects on the expression of genes involved in starch metabolism in wheat grains we would also be able to examine the effects on the tissue specification and organisation in the grain (Hands et al. 2012 J. Exp. Bot. 63,6253) and if this affects fundamental macro characters such as grain shape and crease depth.

### **Interaction of light and gibberellins in the regulation of wheat architecture**

Prof. Matthew Terry, University of Southampton, Prof. Peter Hedden & Dr Steve Thomas, Rothamsted Research

The introduction of reduced height (*rht-1*) gain-of-function mutations that cause constitutive



activity of the growth-repressing DELLA protein RHT-1 into wheat was critical to the success of the Green Revolution, resulting in substantial increases in productivity. We are currently testing the hypothesis that RHT-1 regulates wheat growth via Phytochrome-Interacting Factor (PIF) proteins. In this project we will construct an RNAi line to the wheat homologue of the Arabidopsis PIF4 gene and test whether this line shows an altered response to gibberellins, which control wheat growth via RHT-1.

### **Role of chitin perception and signalling in plant-pathogen interactions**

Kostya Kanyuka, Rothamsted Research

Silencing of wheat pattern recognition receptors CERK1 and CEBiP via a stable RNAi approach will help to elucidate roles of these genes in wheat defence to a range of chitin containing pathogens such as fungi and insects, and indicate whether these genes may serve as good targets for manipulation and engineering crop plants with enhanced resistance to agriculturally important pests and diseases.

### **Using broad-spectrum ABC transporters to probe the wheat basal immune system**

Mike Deeks & Nicholas Talbot, University of Exeter

When activated with sufficient speed and aggression, the plant basal immune response can impede otherwise virulent pathogens. Unlike gene-for-gene resistance, these defences can be broad-spectrum and effective against multiple classes of microbe. Understanding how yield-reducing diseases of cereals evade these defences will lead to new synthetic biology, breeding and priming strategies for crop protection. To achieve this, these dynamic and microscopic molecular mechanisms must be understood at the living plant-pathogen interface. We are proposing to assemble a series of compatible live-cell imaging probes in wheat capable of reporting the earliest and most dynamic events occurring in the wheat basal immune system as it responds to disease-causing

pathogens. Each line can be used effectively in isolation but when combined methodically for dual-label imaging these probes will enable a systems-level oversight of the basal immune response. The CRWT line produced by this project will express a fluorescent protein fusion to a wheat ABC transporter protein; a class of proteins known to confer broad-spectrum microbial resistance. This line will be used as a tool for studying the host basal immune response and mechanisms of defence evasion in multiple wheat-compatible pathogens.

### **Manipulating plastid development in wheat using the SP1 ubiquitin E3 ligase**

Prof. Paul Jarvis, University of Oxford

The development and functions of plastids (e.g., photosynthetic chloroplasts and starch-storing amyloplasts) are governed by the import of thousands of nucleus-encoded proteins from the cytosol. The SP1 E3 ligase controls which proteins are imported into plastids by acting directly on the import machinery (Science 2012, 338:655-659), and so by manipulating SP1 activity we hope to modify plastid development (e.g., amyloplast biogenesis in the grain) in transgenic wheat plants.

### **RNAi-mediated down-regulation of TaMTP8.1 in wheat to investigate mechanisms of Mn homeostasis**

Dr Lorraine E. Williams, University of Southampton

Manganese (Mn) is an essential micronutrient in plant nutrition, yet is detrimental when present in excess. Mn has numerous functions in plants, most notably its role in photosynthesis, operating in photosystem II where it is required to catalyse the oxidation of water, releasing atmospheric oxygen. Mn deficiency and toxicity are widespread agricultural problems, leading to loss of crop yield worldwide. Mechanisms are required to ensure sufficient uptake of Mn into the root and its correct partitioning to plant organs, cells and subcellular organelles. Conversely, Mn must not be allowed to



accumulate where it might have damaging effects. Plants possess a wide range of membrane transporters to alleviate and avoid extremes of Mn nutrition (Williams and Pittman 2010). A key family controlling Mn homeostasis is the Metal Tolerance Proteins (MTPs). Relatively little is known about the mechanism of action of the Mn-MTPs but they are predicted to remove excess Mn from the cytoplasm, either by sequestration into the vacuole, or to the Golgi for vesicular trafficking of Mn to the plasma membrane, and subsequent exocytosis. OsMTP8.1 is important for alleviating Mn toxicity in rice (Chen et al. 2013) and we have recently identified homologues in wheat. This project aims to silence the three TaMTP8.1 homeologues via RNAi to investigate whether this function is conserved in wheat. If so, this transporter could provide a route for improving the tolerance of wheat to Mn nutritional extremes.

#### **Assessing cystatin for stress tolerant wheat**

Prof. Christine Foyer, University of Leeds

Ectopic phytocystatin expression has been used extensively in pest management but the cysteine protease targets of these inhibitors also have important functions in plant biology including lifespan control and stress tolerance. We have previously shown that ectopic expression of the rice cystatin, orzacystatin (OCI) in soybean led to delayed leaf senescence and enhanced tolerance to drought stress, as well as high seed protein accumulation. The aim is therefore to determine whether this technology can be effectively transferred into wheat in order to improve performance and quality traits.

#### **Generation of CAD mutants in wheat**

Prof. Claire Halpin and Prof. Robbie Waugh, University of Dundee at the JHI

High digestibility 'brown-midrib' varieties of maize and sorghum are grown in the USA for animal forage. Some 'brown-midribs' are recessive mutants in a cinnamyl alcohol dehydrogenase (CAD) gene involved in stem lignification. No such phenotype has been

discovered in wheat despite screens for it. TALENs (Transcription Activator-like Effector chimeric Nucleases) have recently been demonstrated to be a novel way of achieving site-specific mutagenesis in plants and could potentially be used to synchronously mutate all three homoeologues of the lignin-related CAD gene in wheat. TALEN cassettes will be designed to simultaneously target all three homoeologues of the major wheat CAD involved in lignin biosynthesis.

#### **Probing endosomal trafficking dynamics using fluorescently labelled ARA6 during infection in wheat**

Dr George R. Littlejohn and Prof. Nicholas J. Talbot, University of Exeter

Wheat blast disease caused by *M. oryzae* Triticum pathotype, emerged in South America in the mid-1980s, and has now expanded its geographic range. It is an important economic and food security threat worldwide as crop yield losses are estimated to be up to 40% in infected areas. Understanding the molecular pathology of this disease is essential for developing effective crop protection strategies. The crucial early stages of infection rely on successful delivery of fungal effector proteins to host targets, allowing the pathogen to evade host defences and re-model plant metabolism in its favour. Endosomal transport is implicated in effector uptake and delivery to structures in the plant cell, including the Biotrophic Interfacial Complex (BIC), a key host-pathogen interface (Giraldo et al. Nature Communications (4), 2013). We will produce wheat lines with fluorescently labelled endosomes to study the infection process.

#### **Engineering nitrogen symbiosis for Africa (ENSA)**

Prof. Giles Oldroyd, JIC

The ENSA project will initiate the first steps towards the transfer of biological nitrogen fixation to cereals, through engineering nodulation signalling. This represents a complex problem. However, the knowledge gained in legumes reveals that much of the machinery



necessary for nodulation signalling is present in cereals and engineering the perception of rhizobial bacteria is likely to be simpler than initially anticipated. It is possible that engineering cereals to perceive the nitrogen-fixing bacteria may allow some degree of plant-bacterial association that could provide some fixed nitrogen without the need for a fully differentiated nodule or the development of complex infection structures. One of the earliest step in this signal transduction pathway is the generation of perinuclear calcium oscillations which are an important precursor to perception of the root by rhizobia. Expression of a calcium responsive synthetic protein, *cameleon*, will allow us to monitor these calcium oscillations and take the first steps towards the transfer of biological nitrogen fixation to cereals, through engineering nodulation signalling in wheat.

#### **Photoreceptor regulation of stomatal development**

Dr Stuart Casson, University of Sheffield

Stomata are pores in the surface of leaves that regulate plant gas exchange. We have demonstrated in *Arabidopsis* that the red-light photoreceptor, phytochrome B, is required for light mediated changes in stomatal development. This project will investigate the impact of altering *phyB* levels in wheat to determine the impact on stomatal development and water use.

#### **Overexpression of RSL genes in wheat to increase yield**

Prof. Liam Dolan, University of Oxford

Root hairs are important for acquisition of nutrients from the soil and seedling establishment. We discovered a class of transcription factors – RSLs – promote root hair development of root hairs in cereals and grasses. Overexpression of RSLs increases root hair development in *Brachypodium distachyon* and *Oryza sativa*. Plants overexpressing these genes also accumulate more biomass and grain than untransformed controls. We propose to generate transgenic wheat that overexpressed a wheat RSL

gene –*TaRSL1*– and determine if it enhances growth.

#### **The role of the TaSPA bZIP transcription factor in wheat grain development**

Dr Sinead Drea, University of Leicester

We propose to characterise the function of *TaSPA*, the orthologue of maize *OPAQUE2* which has been shown to have major effects on quality through its regulation of endosperm-expressed genes such as storage proteins. The *SPA* homeologues will be silenced by RNAi to explicitly characterise the gene function in the wheat grain and assess how it affects expression of candidate target genes in the endosperm and how that further affects the composition of the endosperm and grain development.

#### **A strategy to improve abiotic stress tolerance and grain yield in wheat using a gene from a resurrection plant**

Dr Cara Griffiths, Dr Matthew Paul, Rothamsted Research

Studies of resurrection plants have enabled researchers to elucidate mechanisms allowing these plants to survive extreme environments. A gene isolated from the resurrection grass *Sporobolus stapfianus* has shown potential in *Arabidopsis* to improve biomass and seed yield production, as well as drought tolerance/recovery. It is rare that a single gene can have both these effects. Introducing this enhanced phenotype in wheat could improve crop productivity as well as drought tolerance.

#### **Calcium regulation of photosynthesis and chloroplast function in wheat responding to abiotic stress.**

Prof. Marc Knight, University of Durham

This project will determine the role played by chloroplastic calcium in tuning photosynthesis to environmental conditions to maximise productivity and minimise stress. Transgenic wheat lines expressing the luminescent calcium reporter *aequorin* in the stroma will be used to measure dynamic changes in the second



messenger calcium in intact plants responding to changing abiotic parameters including stress conditions. By measuring and manipulating stromal calcium, we will determine the level and nature of the regulation of photosynthesis by calcium.

#### **Improving silicon uptake in wheat.**

**Dr Frans Maathuis, University of York**

Wheat production suffers great losses to biotic and abiotic stress. Silicon (Si) is indispensable in optimising plant health, especially in cereals, where it improves mechanical properties of many plant parts. In that manner Si increases resistance against pathogens and herbivores, it reduces transpiration and thus drought and salinity stress and it reduces lodging. To increase Si uptake in wheat, we want to overexpress the main uptake system Lsi1 and test how this affects wheat growth and stress tolerance.

#### **Wheat transformation to increase grain yield and industrial quality by improving kernel weight**

**Prof. Simon McQueen-Mason, University of York and Prof. Daniel Calderini, University of Valdivia, Chile**

Kernel expansion precedes grain filling during development and determines final kernel size. The pericarp forms the outer constraining tissue and endosperm the pressure force of developing wheat kernels. Expansins play a key role in growth by inducing the expansion of cell walls, which determines cellular growth rate. We will overexpress expansins in wheat pericarp and endosperm during growth to test the hypothesis that increased pericarp and endosperm growth will lead to larger kernel size, increased yield and improved industrial quality.

#### **Understanding signalling pathways protecting chloroplasts against photo-oxidative stress in wheat through downregulation of a wheat PIF3 orthologue by RNAi**

**Prof. Matthew Terry, University of Southampton**

The introduction of reduced height (rht-1) mutations that cause constitutive activity of the DELLA protein RHT-1 into wheat was critical to the success of the Green Revolution. DELLA proteins have also been shown to protect against oxidative stress at least in part through their interaction with Phytochrome-Interacting Factor (PIF) proteins. In this project we will construct an RNAi line to the wheat PIF3 orthologue and test whether wheat PIF3 is required for RHT-1 mediated responses to photo-oxidative stress.

#### **Misexpression of pollen development genes for control of male fertility**

**Prof. Zoe Wilson, University of Nottingham**

Key advances have been made in understanding pollen biology using the molecular genetic resources available for Arabidopsis to dissect the pollen regulatory gene networks in other species. This project will transfer our understanding of pollen development pathways from Arabidopsis to wheat, by analysis of misexpression of a transcription factor involved in pollen formation. This will aid functional analysis of the gene, but also serve as a potential mechanism for the future, to control fertility in wheat.

#### **Understanding the rooting and germination functions of wheat ARABIDILLO homologues**

**Dr Juliet Coates & Dr Daniel Gibbs, University of Birmingham**

Seed germination and root development are both critical plant developmental processes with key agricultural importance. ARABIDILLO (Arabidopsis Armadillo) genes and their ancient homologues regulate germination in response to abscisic acid, a key plant stress hormone, while also performing a later-evolving function in the precise control of root architecture. This project will define for the first time the functions of cereal ARABIDILLO homologues, and determine whether they can



enable improved crop performance via changes in root architecture and germination responses.

#### **Boosting meiotic crossover in wheat via a natural modifier of recombination**

Dr Charles Underwood, Dr Piotr Ziolkowski and Dr Ian Henderson, University of Cambridge

Boosting meiotic recombination has the potential to accelerate breeding and secure future yield increases. Meiotic recombination in natural plant populations is under genetic control, although causal loci have remained elusive. Using an Arabidopsis QTL mapping population from two divergent accessions (Columbia and Landsberg), we have mapped a natural modifier of recombination rate. We propose to transform the modifier into wheat and test for similar increases in recombination.

#### **Misexpression of pollen development genes for control of male fertility**

Prof. Zoe A Wilson, University of Nottingham

Feeding an increasing population with less resources and land requires a substantial and constant increase in productivity. Heterosis, or hybrid vigour, has been shown to increase yield by 3.5-15%, and has been successfully commercialised in crops such as maize, rice, barley and wheat. This project will reduce the expression of the wheat CSA gene by overexpressing the gene fused to the SRDX transcriptional repression domain to generate transgenic lines with environmental dependant fertility, to be used as a tool in hybrid breeding.

#### **Manipulating chromosome pairing and recombination by silencing wheat CDKG1 kinases**

Prof. John H. Doonan, Aberystwyth University

During meiosis, recombination generates genetic diversity. We need to understand how the recombination process can be altered and tailored to meet the specific needs of different breeding strategies. We recently demonstrated that in Arabidopsis, abolishing expression of CDKG1 dramatically affects pairing and

recombination. We propose to determine if wheat CDKG1 plays a similar role, using an RNAi approach to reduce gene expression. This could define a novel, evolutionary conserved pathway to regulate pairing and recombination in plants.

#### **Generation of CRISPR-Cas9 knock-outs in wheat** Prof. Claire Halpin, University of Dundee

We will exemplify the value of using CRISPR-Cas9 for generating gene knock-outs in wheat. CRISPR-Cas9 is a transformational technology for crop science and breeding. We have used it successfully in diploid barley and now want to exemplify it in hexaploid wheat and evaluate its efficiency to underpin future work. We will target the CAD gene involved in stem lignification to give rise to the first high-digestibility 'brown-midrib' mutant in wheat.

#### **Protecting crop yields through stomatal cooling** Prof. Julie Gray, University of Sheffield

Crop fertility is vulnerable to heat stress and this is set to become an increasing problem under warmer climate scenarios. Wheat pollen and seed set are particularly sensitive to heat stress, and it is predicted that damaging temperatures during the reproductive stage will soon have a major effect on future grain yields. We will investigate whether manipulating plant development to enhance transpiration and evaporative cooling could protect wheat yields at high temperatures.

#### **Enhancing water use efficiency (WUE) and drought tolerance in wheat**

Prof. Marc Knight

We have screened Sorghum bicolor for genes responsible for staygreen drought tolerance/water use efficiency (WUE) traits. One such gene is a close homologue of SDIR1 from Arabidopsis that positively regulates stress-responsive abscisic acid signalling. Overexpression of this Sorghum gene (SbSDIR1) in Arabidopsis led to reduced transpiration, with no penalty on growth/yield, as well as enhanced drought tolerance. Thus this protein represents an excellent candidate for the development of



wheat strains with increased water-use efficiency and/or drought tolerance.

### **A non-invasive luciferase reporter for measuring circadian rhythms in wheat**

Prof. Alex Webb, University of Cambridge

Understanding the role of the circadian clock in wheat has been limited by the lack of a non-invasive reporter of circadian rhythms. We will make a transcriptional fusion of the promoter the circadian clock gene TaTIMING OF CAB1 (TOC1) fused to firefly luciferase (LUC+) to measure circadian rhythms by photon counting imaging. This can be crossed with any hexaploid wheat background to investigate circadian clock function in varieties with suspected lesions in the circadian system.

### **Investigating the role of the N-end rule pathway E3 ligase, TaPRT1 in agronomically-important traits**

Prof. Frederica Theodoulou, Rothamsted Research and Prof. Michael Holdsworth, University of Nottingham

The N-end rule pathway is a specialised branch of targeted protein degradation which links the fate of a protein to its amino- (N-) terminus. PROTEOLYSIS1 (PRT1) is an E3 ubiquitin ligase that recognises aromatic N-termini but until recently, its physiological functions remained elusive. Recently, we have obtained evidence for key roles for Arabidopsis PRT1 in agronomically relevant processes. This project seeks to test whether the function(s) of PRT1 are conserved between wheat and Arabidopsis.

### **Novel source of broad-spectrum disease resistance derived from elite wheat cultivar**

Dr Ksenia Krasileva, The Genome Analysis Centre and The Sainsbury Laboratory

We have screened Targeted Induced Lesions in Genomes (TILLING) population of elite durum wheat *Triticum turgidum* cv. Kronos for induced allelic variation confirming gain-of-resistance against threatening wheat pathogen, yellow rust. We have identified novel alleles of the resistance

gene that we hypothesize will provide broad range resistance to multiple wheat pathogens. We are proposing to validate this novel candidate allele in bread wheat by transgenic approach.

### **Engineering freezing and drought tolerance in wheat**

Prof. Marc Knight, Durham University

We have screened the monocot crop *Sorghum bicolor* for genes responsible for “staygreen” drought tolerance/water use efficiency (WUE) traits. One such gene is a close homologue of DREB1 from Arabidopsis which is a transcription factor that induces cold and drought tolerance genes. Overexpression of DREB1 in Arabidopsis leads to both enhanced freezing and drought tolerance (Jaglo-Ottosen et al., 1998). Thus this protein represents an excellent candidate for the development of wheat strains with increased freezing and drought tolerance.

### **Investigating the role of TaMADS47 in regulating prolamin storage proteins in wheat**

Dr Sofia Kourmpetli, Cranfield University & Dr Sinéad Drea, University of Leicester

The transcriptional regulation of grain storage proteins in wheat is still not well understood. ZmMADS47, a MADS-box protein, has recently been shown to interact with OPAQUE2 (O2) and regulate the transcription of zein storage proteins in maize endosperm. We have identified the wheat orthologue of ZmMADS47, TaMADS47, and by generating RNAi transgenic lines we will be able to understand its role in wheat prolamin regulation and shed more light into the complex genetic networks that control wheat grain quality.

### **Rewiring photosynthesis with a heterologous electron sink to enhance photoprotection in wheat**

Dr Thomas S Bibby, University of Southampton

The majority of light absorbed by photosynthetic species is lost before it can be stored in chemical-bonds. Our objective is to reengineer photosynthesis such that lost energy is diverted





to drive useful chemical reactions. We will rewire photosynthesis in wheat, by installing a cytochrome P450 into the photosynthetic electron transport chain as an artificial electron sink. This will increase both the light intensity at which photosynthesis is saturated and catalyse the degradation of pollutants/herbicides.

### **Improving wheat by manipulating stomatal responses to CO<sub>2</sub>**

Prof. Alistair Hetherington, University of Bristol

Stomatal aperture decreases in response to increased atmospheric CO<sub>2</sub>. In the absence of other compensatory factors this will impact negatively on wheat grain mineral nutrient content. To mitigate this effect we will make wheat plants that will not exhibit reductions in stomatal aperture in response to growth at increased atmospheric CO<sub>2</sub>. We will measure water use efficiency, yield and grain Fe and Zn in these plants at ambient and elevated CO<sub>2</sub> and compare it with Fielder.

### **Improving organic nitrogen uptake in wheat**

Dr Chris West, University of Leeds

Nitrogen is often growth limiting for crops and supplemental nitrogen-based fertilisers are required for optimal yields. The energy costs in chemical N-fixation is high and heavily reliant on fossil fuels. Increased usage of organic sources, and improved organic N acquisition by wheat provides a sustainable route to reduced inorganic nitrogen whilst maintaining yield. Modification of wheat to promote uptake of peptides from the soil will improve the efficiency of organic fertilisers and nitrogen acquisition from poor soils of higher organic nitrogen content.

### **Monitoring synaptonemal complex biogenesis in vivo utilising a fluorescently tagged wheat ZYP1 protein**

Dr James Higgins and Dr Stuart Desjardins, University of Leicester

The hexaploid bread wheat genome has arisen through two independent hybridisation events between three diploid progenitors. During

meiosis, a specialized cell division that leads to the production of pollen and ovules, the correct homoeologous chromosomes must pair and recombine to avoid genome instability and chromosome mis-segregation. We aim to understand this important and complex process by monitoring synaptonemal complex formation (a structure that physically links homoeologous chromosomes and prevents recombination defects) in vivo with a fluorescently tagged ZYP1 protein.

### **Betaine synthase from diatoms - engineering more nutritional and salt tolerant crops**

Ana Bermejo Martinez and Dr Jonathan Todd, University of East Anglia

Glycine betaine is a well-known osmoprotectant whose accumulation in the cells has been shown to increase salt tolerance in many plants. The single enzyme for glycine betaine synthesis, identified from marine diatoms, is a new tool to accumulate this osmolyte more efficiently in the plants. Salt tolerant crops are of great interest as saline agricultural soils are an increasing problem around the world.

### **Developing mitochondrial mediated resilience to abiotic stress in wheat**

Prof. Zoe A Wilson, University of Nottingham

The mitochondrial uncoupling proteins (UCPs) are among the ~1,000 proteins that compose the mitochondrial architecture and play a key role in securing integrity under stress. We have demonstrated that the constitutive overexpression of UCP1 induces a broad metabolic reconfiguration in the cellular context and makes plants more tolerant to several biotic and abiotic stresses. This project aims to explore the effect of UCP1 overexpression in wheat, to establish if this confers enhanced resilience to stressful environments during growth.



## **Generating steeper and deeper rooting wheat lines**

Dr. Stefan Kepinski, University of Leeds

The modification of root system architecture is predicted to be a key component in the development of crops that perform well in lower input agricultural systems and on dry, infertile soils. We have developed a technology to alter root growth angles by the targeted manipulation of auxin signalling in the gravity-sensing cells of the root. Here we propose to trial this technology in wheat with the aim of generating lines with steeper, deeper root systems.