We seek to identify the geographic regions that best explain the diversity in domesticated barley and associate genes or genomic regions with diverse environmental conditions.

Molecular tools and genetic resources for eyespot resistance in wheat.

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Eyespot is an important stem base disease of wheat caused by two species of fungi, *Oculimacula yallundae* and *Oculimacula acuformis*, both of which are widespread in northern Europe and the Pacific northwest USA. There are two main sources of resistance available; *Pch1*, a potent resistance gene introduced from *Aegilops ventricosa*, and the moderate resistance of the variety Cappelle Desprez.

Pch1 has not been used widely in European wheat varieties due to linkage between *Pch1* and yield-limiting genes. Breaking this linkage has proven difficult because of a low recombination frequency and a lack of molecular markers that function in both wheat and *Ae. ventricosa*. Utilising the *Brachypodium* genome sequence, we developed co-dominant Conserved Orthologous Sequence (COS) markers targeted to the *Pch1* region. These markers were used to identify recombinants and to locate *Pch1* on chromosome 7DL. By exploiting co-linearity between *Brachypodium*, rice and sorghum, we identified a candidate gene region as a prelude to the map-based cloning of the gene.

Resistance in Cappelle Desprez has been attributed to the partial resistance gene *Pch2*. We mapped *Pch2* resistance to *O. acuformis* as a QTL on the distal end of chromosome 7AL. However, it was not possible identify any resistance to *O. yallundae*. This indicates that *Pch2* operates specifically against *O. acuformis*, and that *Pch2* should not be relied upon as a stand-alone resistance in wheat varieties.

There is previous evidence of an adult-plant resistance on chromosome 5A of Cappelle Desprez. We showed this resistance to also be effective in seedlings and to provide protection against both pathogen species, indicating that it will be useful in commercial varieties. Two chromosome 5A recombinant populations were tested in seedling bioassays and field trials to locate a stable resistance QTL on 5AL and identify linked SSR markers for selection of the resistance.

In addition to characterising existing eyespot resistances, we screened 1,036 hexaploid genotypes from the Watkin's Worldwide Collection for resistance to *O. yallundae*. Some accessions exhibited a high level of resistance. Selected lines will be screened for resistance to *O. acuformis* and the genetic basis of any novel resistances will be determined.

Wheat Zapper, Illumina BSA, and radiation hybrids for synteny analysis of the scs region.

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Species cytoplasm specific (*scs*) genes are of great evolutionary importance because they guide the precise interaction between nucleus and cytoplasm. Recently, an *scs* gene of wheat (*Triticum* species) was localized to the pericentromere of chromosome 1DL. In an attempt to clone this gene, we generated a radiation hybrid (RH) *in vivo* population of ~1,500 individuals. EST-based markers were developed and used for genotyping the whole 1D chromosome. An online tool was developed to easily and rapidly study the colinearity between wheat and three sequenced plant species (*Sorghum bicolor*, *Brachypodium distachyon*, and *Oryza sativa*), namely the Wheat Zapper (http://wge.ndsu.nodak.edu/wheatzapper/). Employing this tool, together with functional analysis of the RH lines, we determined that the *scs* region corresponds to a region of paleofusion between two ancestral chromosomes corresponding to the modern Sb9-1, Bd3-2, or Os10-5. To

further investigate the events that lead to this fusion, we coupled a protocol for genome complexity reduction with the Illumina platform, to sequence at 40X coverage a bulk of seven RH lines carrying or missing the *scs* gene. The sequences identified in the first (positive) bulk but not in the second (negative), were considered to represent the *scs* region. These 3.2 K differential pair-ends sequences were then extended to 1–5 Kb in size, employing the publicly available 50X survey sequences of *Aegilops tauschii*. Furthermotr, these extended sequences were assembled with DNAStar into 556 contigs spanning 1.2 Mb with an N50 of 3Kb. This gapped sequence was then anchored to the scaffold RH map, pin-pointing the *scs* locus to an interval of just eight genes. The result of coupling Illumina sequencing, Wheat Zapper synteny analysis, and RH populations is discussed here in relation to the evolutionary importance of the *scs* region and its map based cloning.

Identification of a candidate barley stem rust susceptibility gene determining the recessive nature of Rpg4-mediated Ug99 resistance in barley.

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The *rpg4*/*Rpg5* locus in barley (*Hordeum vulgare*) provides recessive resistance against several wheat stem rust races (*Puccinia graminis* f. sp. *tritici*) including QCCJ and the highly virulent race TTKSK (aka Ug99). Three genes required for wheat stem rust resistance (*HvADF3*, *Rpg5*, and *HvRGA1*) were identified in the ~70kbp *rpg4*/*Rpg5* stem rust resistance locus using high-resolution mapping and virus induced gene silencing. The dominant rye stem rust resistance gene *Rpg5* is predicted to have the typical R-gene domains including the nucleotide-binding site (NBS), leucine rich repeat (LRR), and serine/ threonine protein kinase (STPK) domains. The *Rpg5* gene appears to condition compatible or incompatible interactions with the wheat stem rust races QCCJ and Ug99, because it is the only polymorphic gene correlating with resistance and susceptibility in the delimited *rpg4*/*Rpg5* region. Sequence analysis of *rpg5* susceptible alleles showed that they make up two groups. The group-1 susceptible lines contain an insertion/deletion region having a predicted functional protein phosphatase 2C gene (*HvPP2C*) in place of the *Rpg5* STPK domain (Harrington, Steptoe, and Sm89010). The group-2 susceptible lines have an intact STPK domain but a predicted nonfunctional *rpg5* allele due to a single cytosine insertion causing a frame shift mutation resulting in a premature stop codon (Golden Promise, OSU6, and MD2). Analysis of F_2 progeny from crosses between Q21861 and group-1 and -2 susceptible lines segregated in 1:3 ratio (resistant:susceptible) when the *HvPP2C* gene is present but in a 3:1 ratio (resistant:susceptible) when the *HvPP2C* gene is absent. Thus, it appears that the previously identified *Rpg5* dominant rye stem rust resistance gene also imparts *rpg4*-mediated wheat stem rust resistance and behaves as a dominant gene in the absence of the *HvPP2C* gene but as a recessive resistance gene in the presence of *HvPP2C*. The data suggests that components of *rpg4* and *Rpg5* resistance are not distinct and the difference in the dominant or recessive nature of resistance is due to the *HvPP2C* gene acting as a dominant susceptibility factor that suppresses *rpg4*/*Rpg5*-mediated resistance against the wheat stem rust races including Ug99.

Targeted re-sequencing of the wheat exome and the generation of public co-dominant single nucleotide polymorphism markers.

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The complex nature of the wheat genome has, until recently, resulted in a lack of single nucleotide polymorphism (SNP)-based molecular markers of practical use to wheat breeders. Recently, large numbers of SNP-based wheat markers have been made available via the use of next generation sequencing combined with a variety of genotyping platforms. However, many of these markers and platforms have difficulty distinguishing between heterozygote and homozygote indi-