



WHAT IF ALL VARIANTS ARE RARE?

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Figure 1. Wheat TILLING panel in the wheat field. Scanalyzer (Vialé *et al.* 2010).



MATERIALS AND METHODS

- We performed quantitative genomic analyses (see pipeline) in an experimental TILLING population of hexaploid wheat cv. Cadenza (~145 lines) in which all chemically induced mutations were either rare or unique, starting with a (mostly) uniform genomic background.
- The field trials were conducted in John Innes Centre (JIC) and at the Field Scanalyzer phenotyping platform at Rothamsted Research, Harpenden, UK.
- Using exome re-sequencing data, we partitioned the complete set of ~7M mutations (~7K genes) into non-synonymous, synonymous, and non-coding sets.
- We conducted gene-based analysis (collapsing models) and variance component analyses for three phenotypic traits (i.e. plant height, heading/flowering time, and senescence).

GENE SCANNING AND VARIANCE PARTITIONING

- Gene scanning of variants revealed association signals coming mostly from *non-synonymous* and *synonymous* substitutions for plant height (Fig. 3).
- Breaking down the genetic variance of chromosomes and single genes revealed insights of the contribution of *ultra-rare* variants to phenotypic traits (Fig. 3).

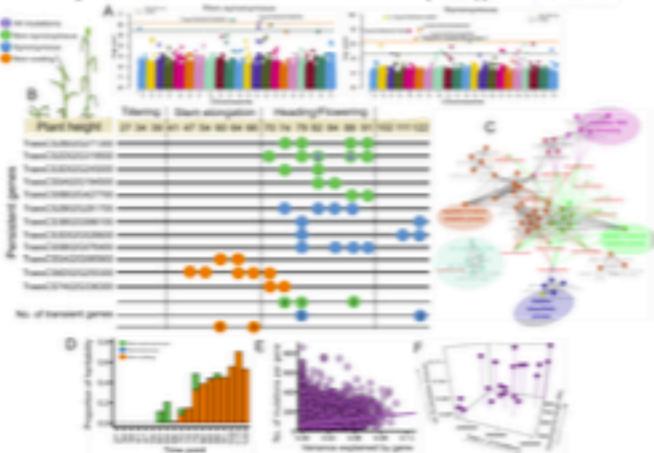


Figure 2. Variance of the quantitative trait of plant height in the wheat TILLING population. (A) Whole genome scanning for plant height mapping in the 100 lines of the wheat TILLING population. (B) Number of mutated genes per line. (C) Network diagram of gene interactions with the highest degree of connectivity. (D) Mutation rate. (E) Mutation rate vs. gene length. (F) Distribution of mutation rates. (G) Allele frequency distribution of mutated alleles of each mutation type for plant height. (H) Correlation between other trait (HL) and MAF in each mutation for plant height.

INTRODUCTION

- TILLING (T_{AGT} Insert L_{AGT} Insert L_{AGT} In G_{AGT}) is a method used to create a *mutagenised* population allowing the identification of point mutations within genes of interest (Krasileva *et al.* 2017).
- The relative contribution of (*ultra*) *rare* variants to quantitative genetic variation is largely unclear, despite recent advances in genomics. One reason for this is that natural populations tend to violate the assumptions of genome-wide association and genomic heritability models.
- Field-based phenotyping tools have been used to identify genes/QTL throughout the crop growth cycle for dynamic phenotypes (Lynn *et al.* 2020).

MAIN OBJECTIVES

The goals of this study were to:

- perform gene-based association analysis using collapsing models into non-synonymous, synonymous, and non-coding mutation sets,
- partition the variance of (a) mutation types over the course of the growing season, (b) single genes, and (c) chromosomes.

QUANTITATIVE GENETICS PIPELINE



ULTRA-RARE VARIANTS IN THE WHEAT TILLING POPULATION

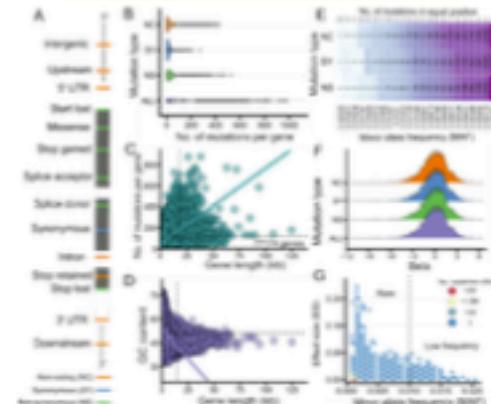


Figure 3. Mutation variant summary in the wheat TILLING population. (A) Schematic diagram of gene clustering of all the mutated genes with the most non-synonymous (NS), synonymous (SYN), and nucleotide (NT) mutations. (B) Total number of mutated genes per trait (plant height, heading/flowering time, and senescence). (C) Number of mutated genes per trait. (D) Number of mutated genes per trait. (E) Number of mutated genes per trait. (F) Number of mutated genes per trait. (G) Number of mutated genes per trait. (H) Correlation between other trait (HL) and MAF in each mutation for plant height. (I) Correlation between other trait (HL) and MAF in each mutation for plant height.

CONCLUSION

This approach will lead to the identification of candidate genes across a range of phenotypic traits and contribute to the development of novel analytical methodology, ultimately enhancing our understanding of the importance of (*ultra*) *rare* variants in both natural and experimental populations.

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