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## Introduction

- Low temperature tolerance (LTT) - an important component of winter hardiness in barley.
- Three major QTLs identified for LTT - mechanisms not fully understood.
- Reported QTLs - *FR-H1* and *FR-H2* ~ 30 cM apart on chromosome 5H and *FR-H3* on the short arm of chromosome 1H.
- FR-H1* cosegregates with *VRN-H1*.
- FR-H2* locus consists of two physically linked clusters of C-repeat binding factor (*CBF*) genes.
- FR-H1*, *FR-H2*, and *FR-H3* characterized in bi-parental mapping populations.
- Vernalization (VRN) – a low temperature response trigger for vegetative-to-reproductive transition.
- Three major QTLs identified for VRN – controlling growth habits (spring, facultative, & winter).
- Reported QTLs – *VRN-H1* on chromosome 5H, *VRN-H2* on chromosome 4H, and *VRN-H3* on chromosome 7H.
- Facultative growth habit defined as spring allele at *VRN-H2* & winter alleles at LTT-related loci and *PPD-H2*.

## Objectives

- 1) Identify novel LTT & VRN QTL(s).
- 2) Identify novel alleles at known LTT & VRN QTL(s).
- 3) Estimate effects and interactions in a large and diverse panel of germplasm.

## Material & Methods

- The LTT panel - 941 accessions with low temperature tolerance selected by 21 barley breeders around the world.
- The panel includes 300 lines from the TCAP Winter-Facultative 6 row set, and winter lines from the AGOUEB and ExBarDiv Projects.
- The panel genotyped with the Illumina Infinium iSelect 9k chip.
- LTT - Planted 2013/14 (14 locations) & 2014/15 (12 locations): Alberta (Canada), Oregon, Idaho, Nebraska, Ohio, Minnesota (USA), France (2 locations), Germany (2 locations), Spain, Hungary, Scotland (UK), Japan.
- VRN – Planted in greenhouse under non-vernalized conditions to measure max days-to-flowering.
- The experimental design - Type II Modified Augmented Design with 5 replicated checks: **Alba** (winter), **Maja** (facultative), **Full Pint** (spring), **winter wheat**, and a **local cold tolerant check**.

## Future

- Further characterization of best accessions - phenotypically and genotypically.
- Identify candidate genes at novel LTT QTLs.
- LTT and VRN allele introgression into adapted germplasm.

## Results

- Differential survival data obtained from 2014 & 2015 Idaho, Minnesota, Ohio (USA), Alberta (Canada), and 2015 Germany (BLL), Hungary, & Nebraska. Otherwise complete survival or minimal differential survival, except Nebraska 2014, where there was 0 survival.
- 41/941 potential facultative accessions based on 2015 greenhouse days-to-flowering (46-96 DTF) and LTT (>50%) (Figures 1, 2, 3 & 4).
- Normal distribution of winter survival (%) across 2014 & 2015 differential environments (Figure 2) with 114/941 accessions with >75% survival.
- No correlation between spike type (2-row vs. 6-row) and LTT (Figure 5).
- FR-H1* & *FR-H2* are the largest effect and consistent LTT QTLs (Figure 7).
- VRN-1* & *VRN-2* are the largest effect and consistent VRN QTLs (Figure 6 & 7).
- Potentially novel QTLs to explore and characterize.

## Discussion

- The panel contains non-VRN germplasm with excellent LTT: useful starting points for breeding.
- Encouraging prospects for maintaining, improving LTT.

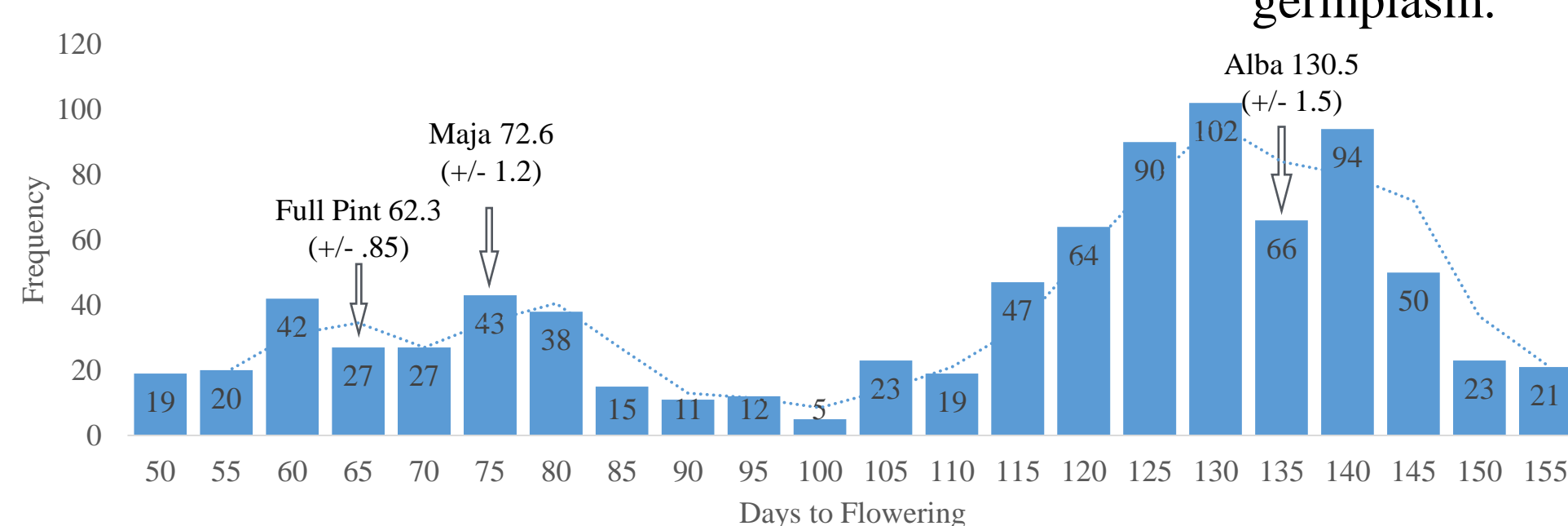


Figure 1. Frequency distribution of 2015 greenhouse days-to-flowering/vernalization sensitivity data of the LTT GWAS panel.

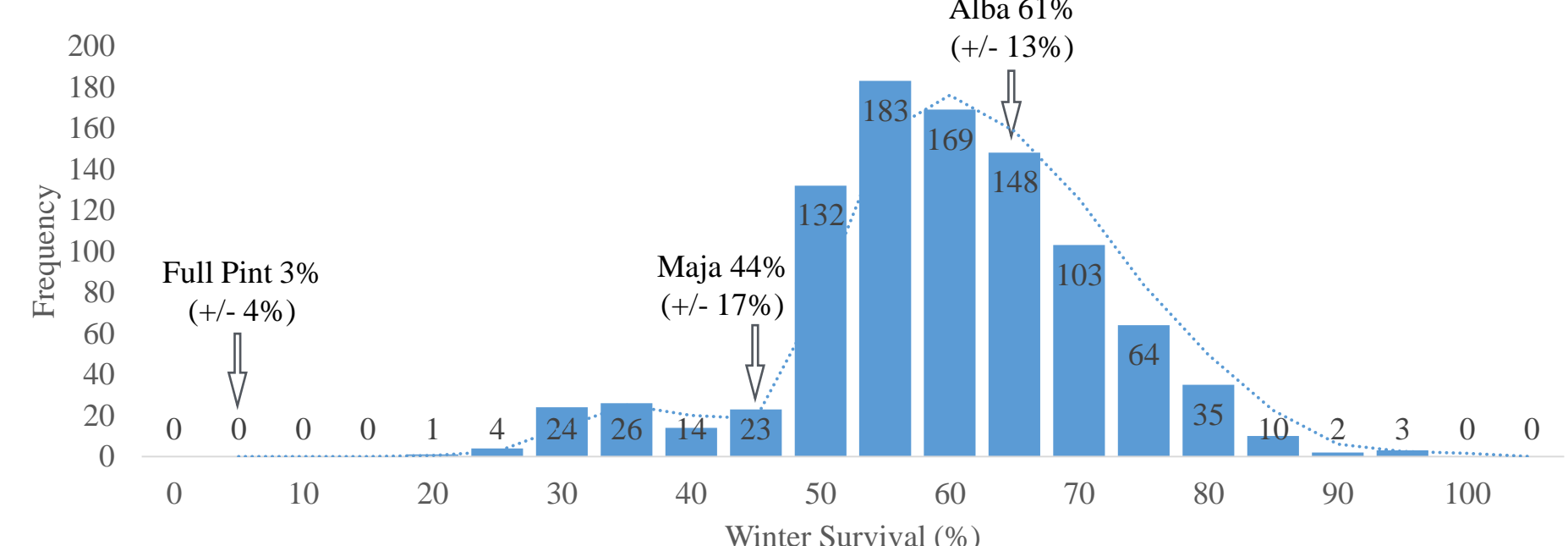
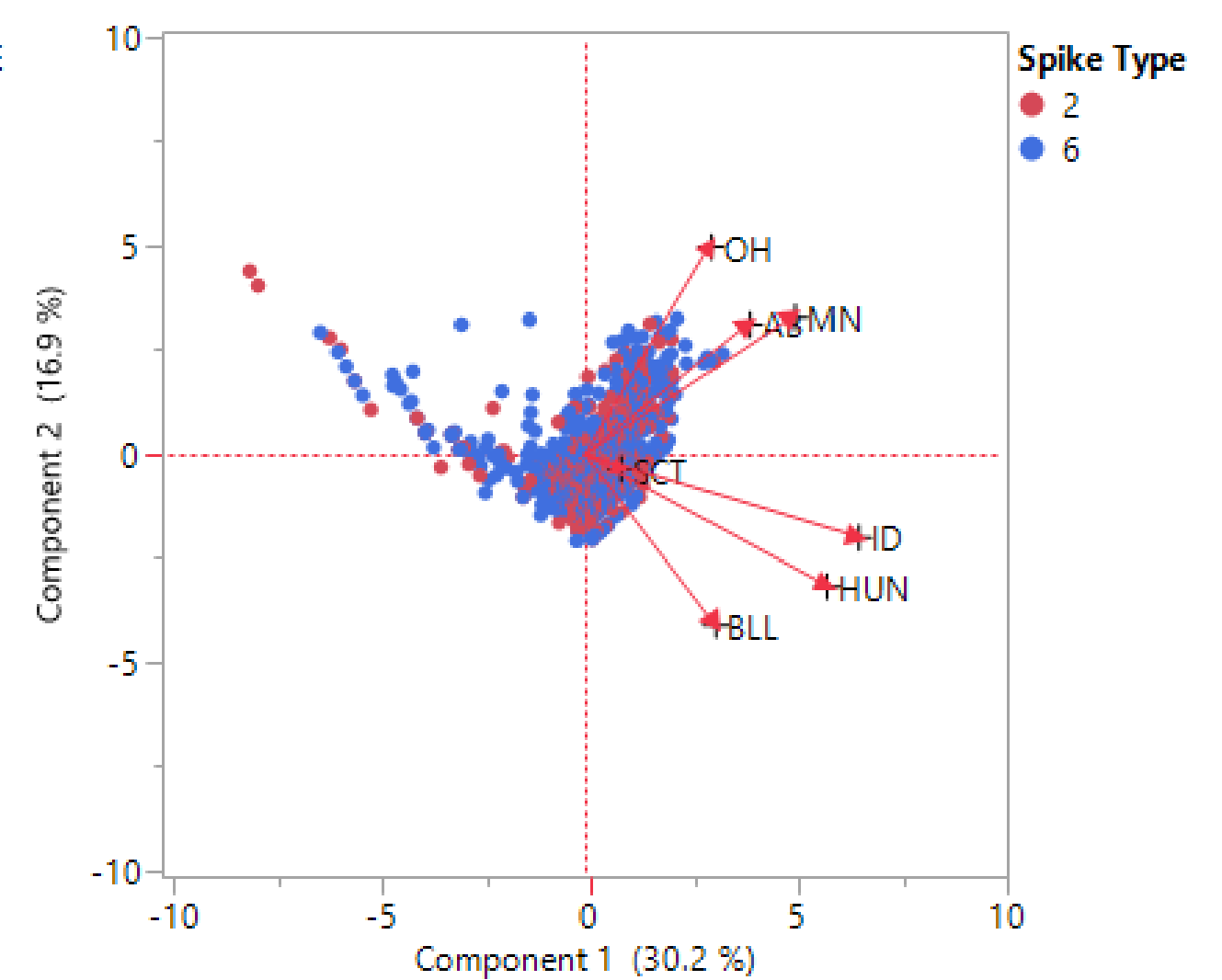
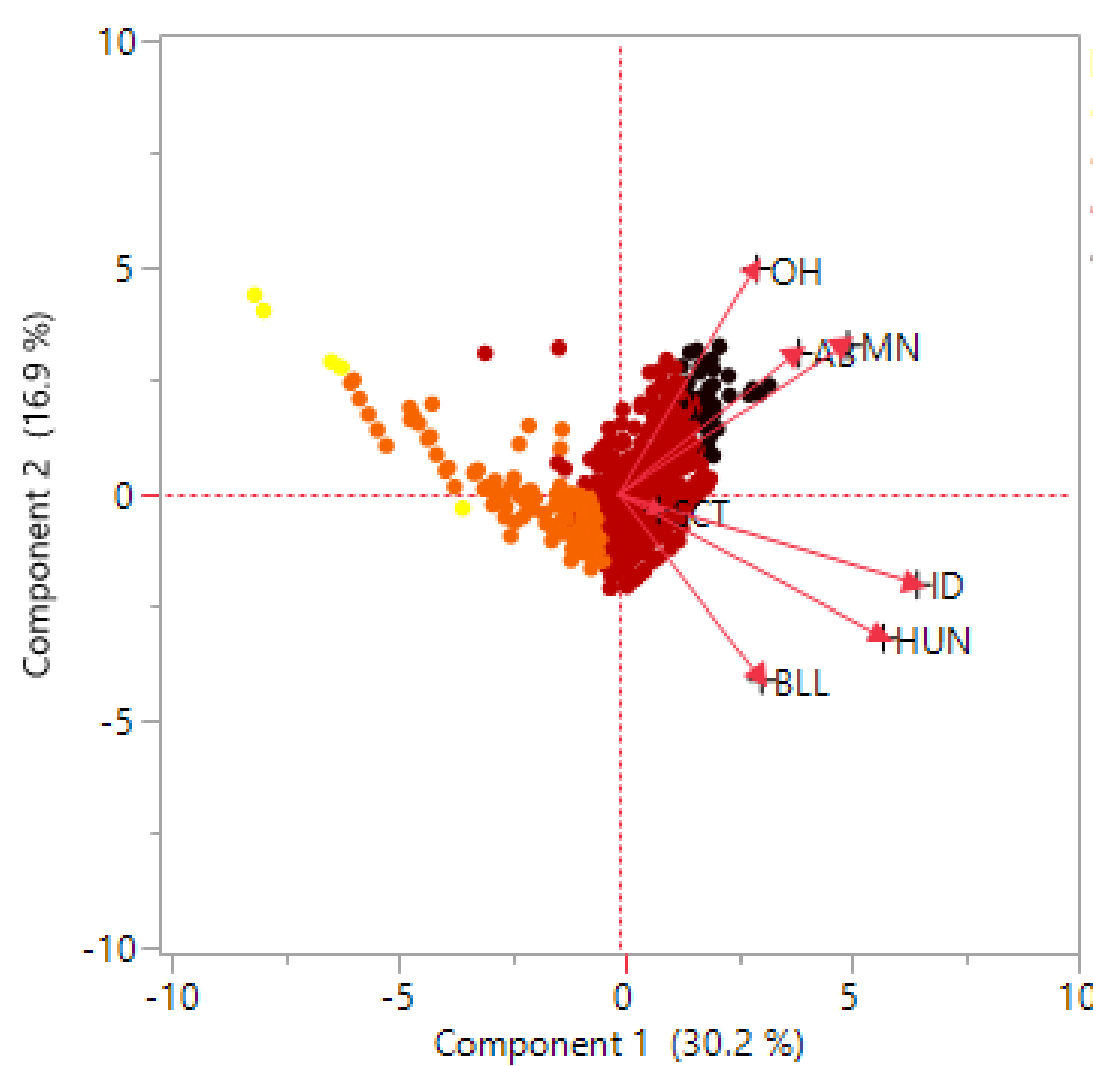
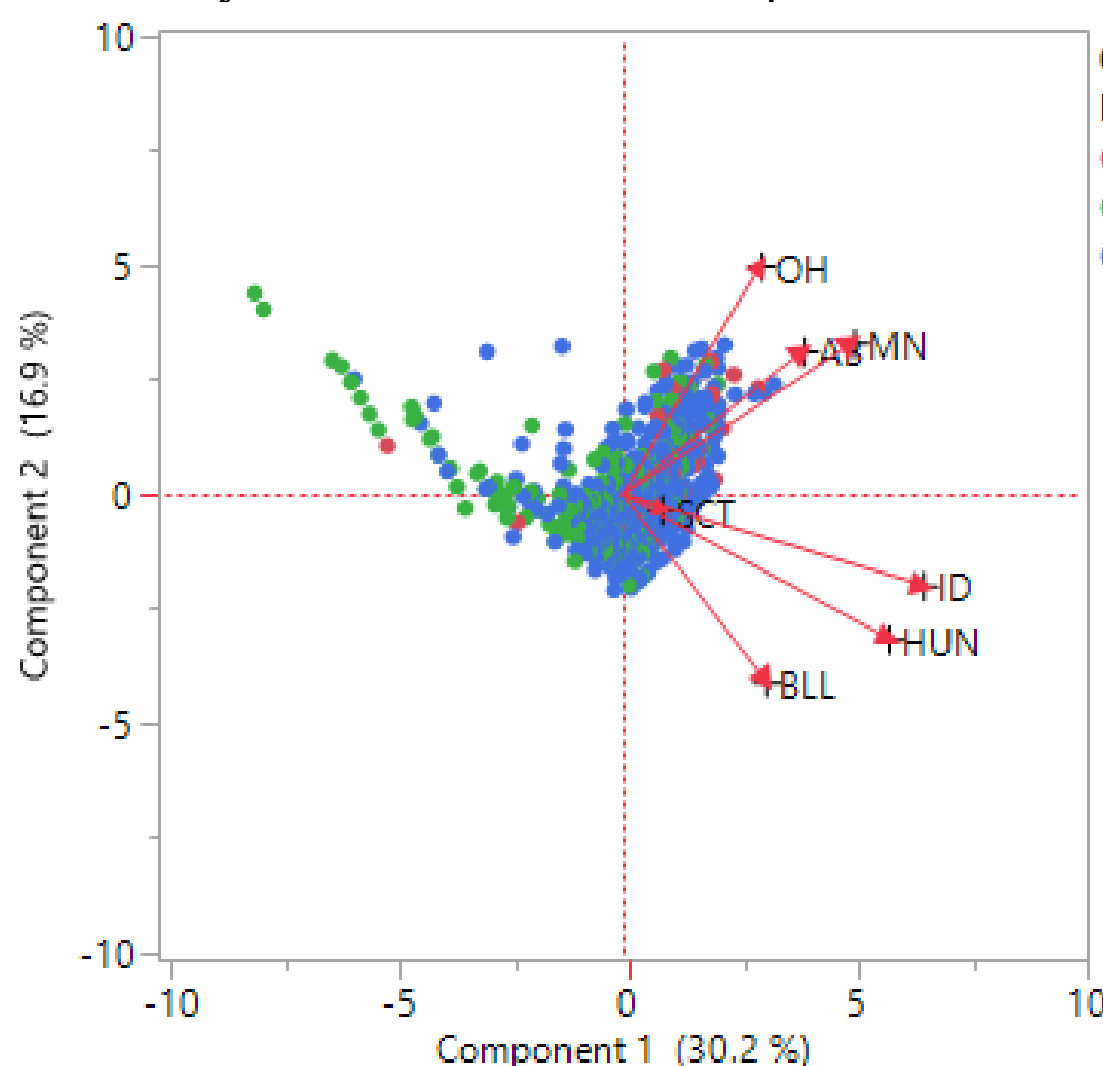


Figure 2. Frequency distribution of average 2014 and 2015 winter survival (%) of the LTT GWAS Panel in differential environments.



Figures 3-5. Principle component analysis of (3) 2015 greenhouse days-to-flowering/growth habit data (Facultative: DTF= 46-96 days & LTT=>75%), (4) combined 2014 and 2015 environments with differential low temperature tolerance data (1= 0-25%; 2= 26-50%; 3= 51-75%; 4= 76-100%), and (5) Spike type (2-row & 6-row).

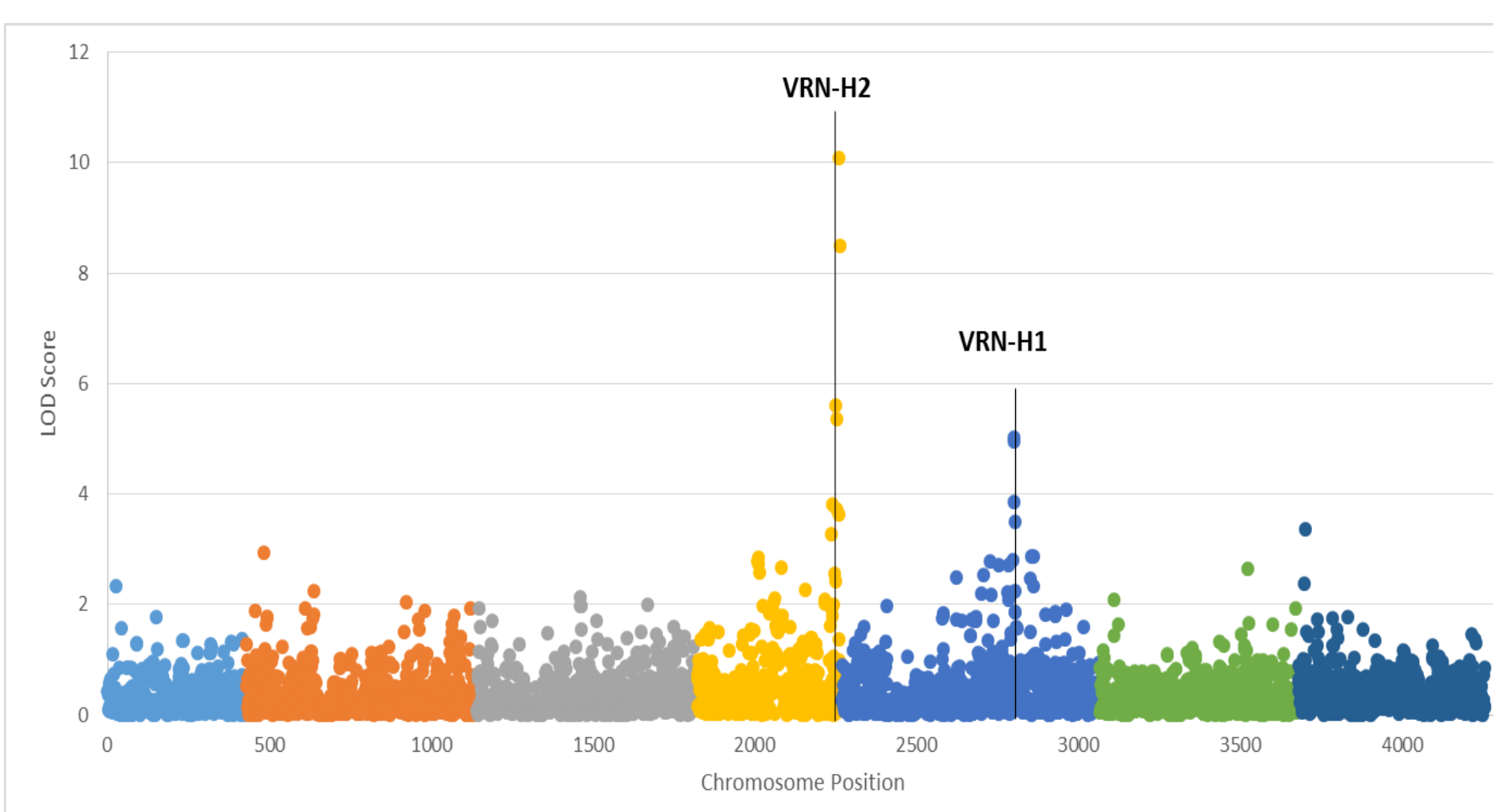


Figure 6. Manhattan plot of marker: trait associations from genome-wide association of 2015 greenhouse days-to-flowering/vernalization data of the LTT GWAS panel.

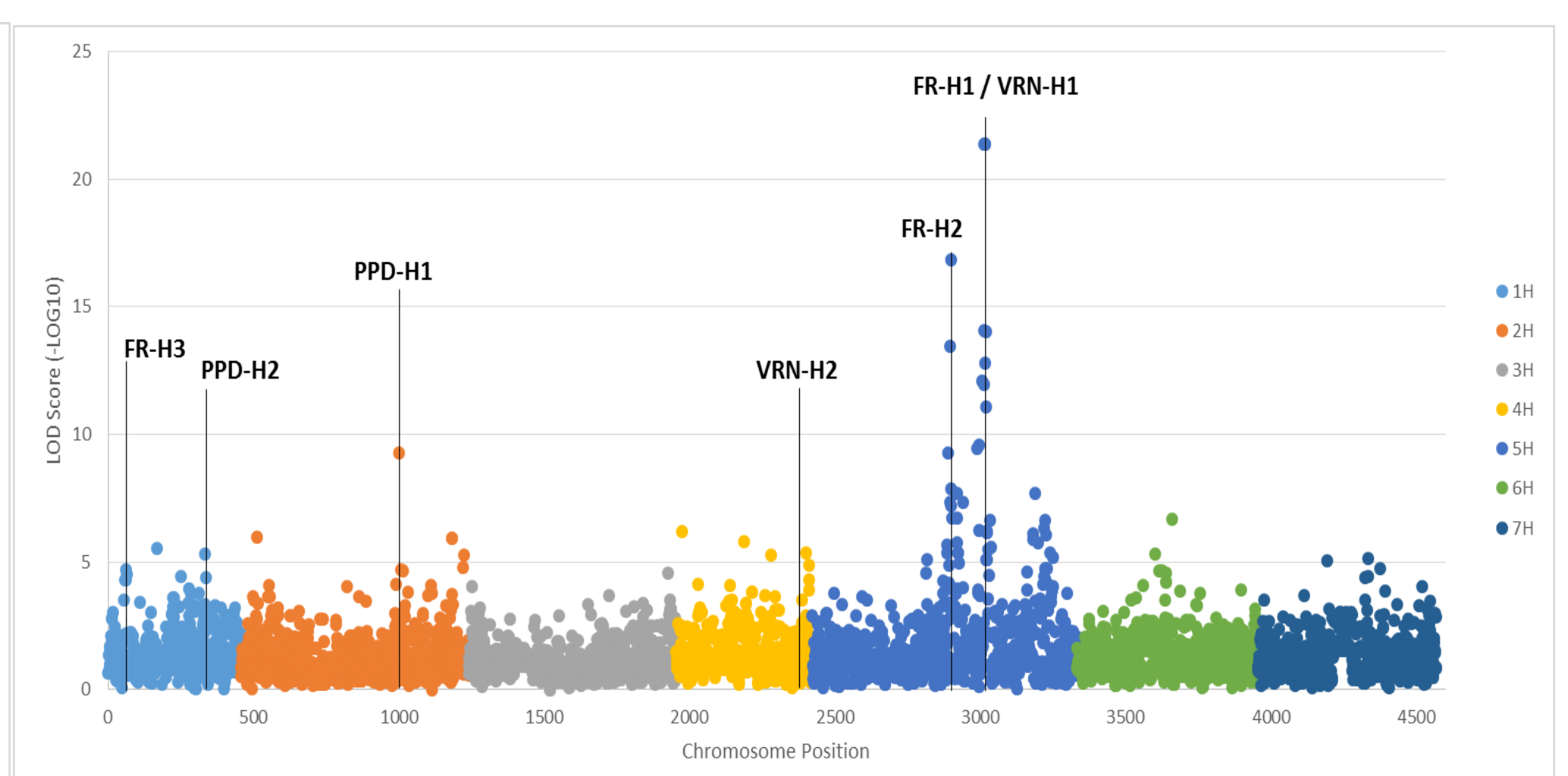


Figure 7. Manhattan plot of marker: trait associations from genome-wide mapping of combined 2014 and 2015 environments winter survival (%) of the LTT GWAS panel.

## Acknowledgements

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