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## Background

Zoysiagrass (*Zoysia* spp.) is one of the most freeze-tolerant warm season grasses. However, there is opportunity for improvement → to compete with cool season grasses north of the transition zone.

Desirable characteristics:

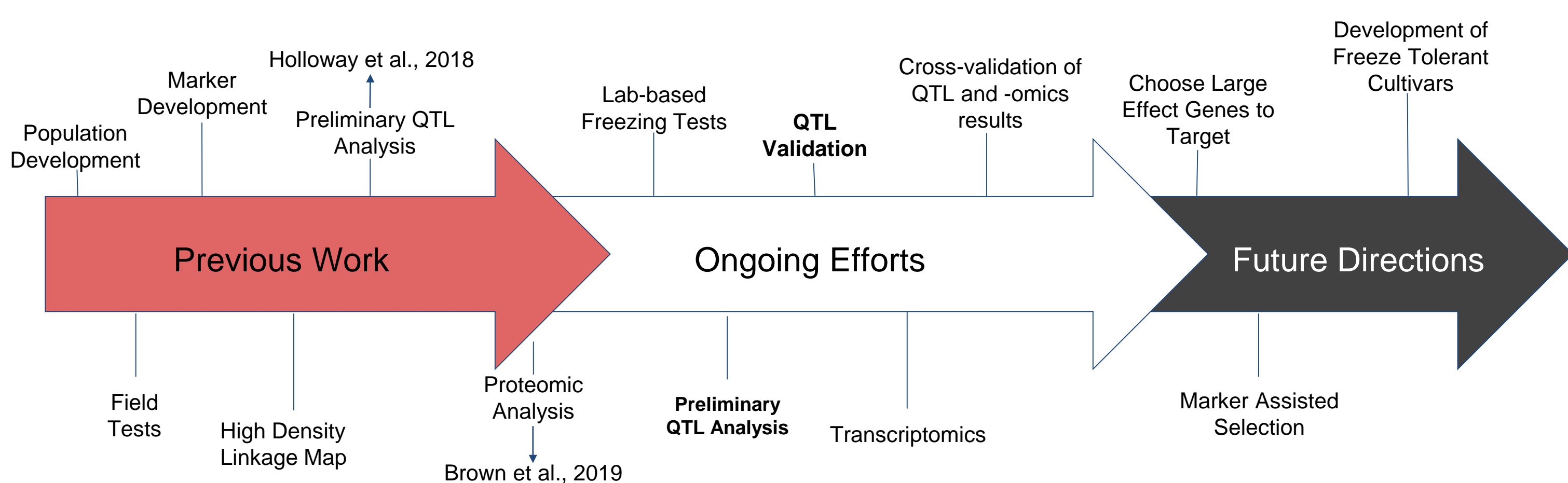
- Dense texture, competitive with weeds, low input requirements, general tolerance to drought, shade and salinity stress.

Previous research by Holloway et al. (2018)

- Generated first high density linkage based map of *Z. japonica*
- Conducted winter field trials at three locations. Identified putative QTL:
  - Seven regions of interest identified on chromosomes 8, 11, and 13
  - Colocation of QTL for three or more traits

## Objective:

- Map QTL for freeze response under controlled environmental conditions in a pseudo-F<sub>2</sub> population of Meyer (freeze tolerant) x Victoria (freeze susceptible).



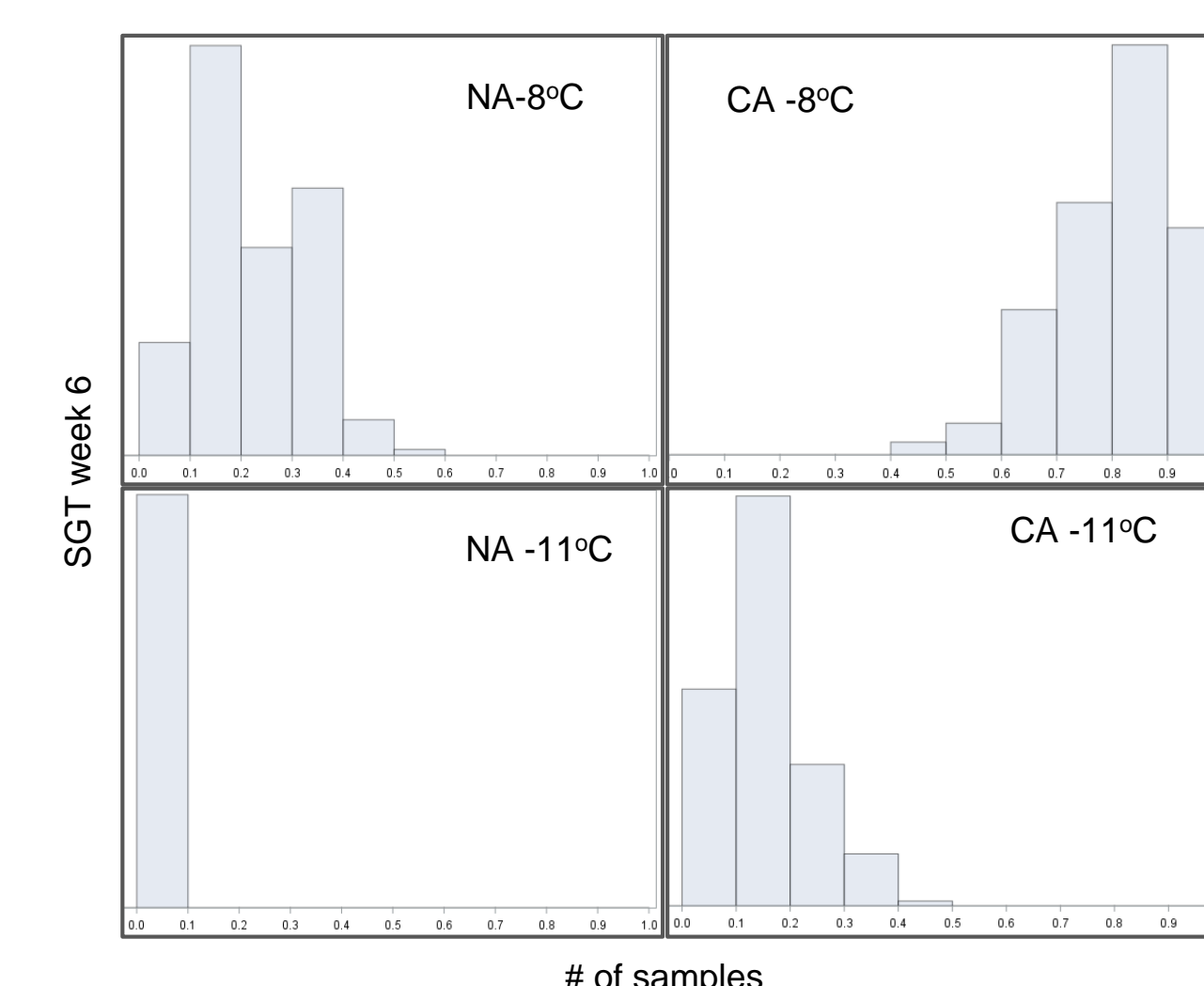
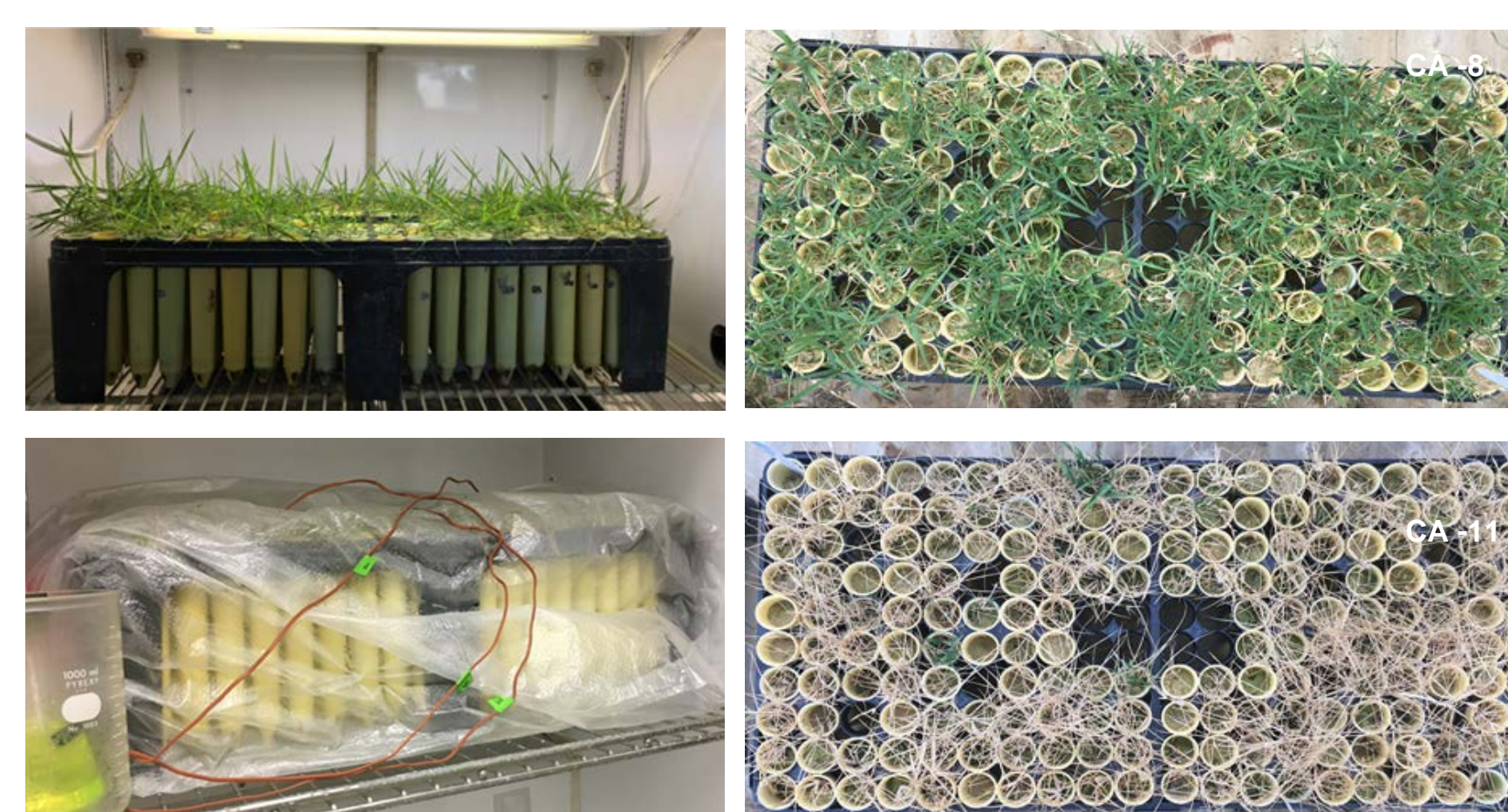
## Materials and Methods

### Freezing Tests

- Evaluate 175 individuals from the mapping population plus the two parents.
- Experimental Design: RCBD with two acclimation (NCA=no acclimation, CA= acclimated 4 wk at 8°C) and two temperature (-8 and -11C °C) treatments.
- Cold acclimation = critical in development of freeze tolerance.
- Rate surviving green tissue (SGT) and regrowth (RG) on 0-5 scale for 6 weeks post-freeze

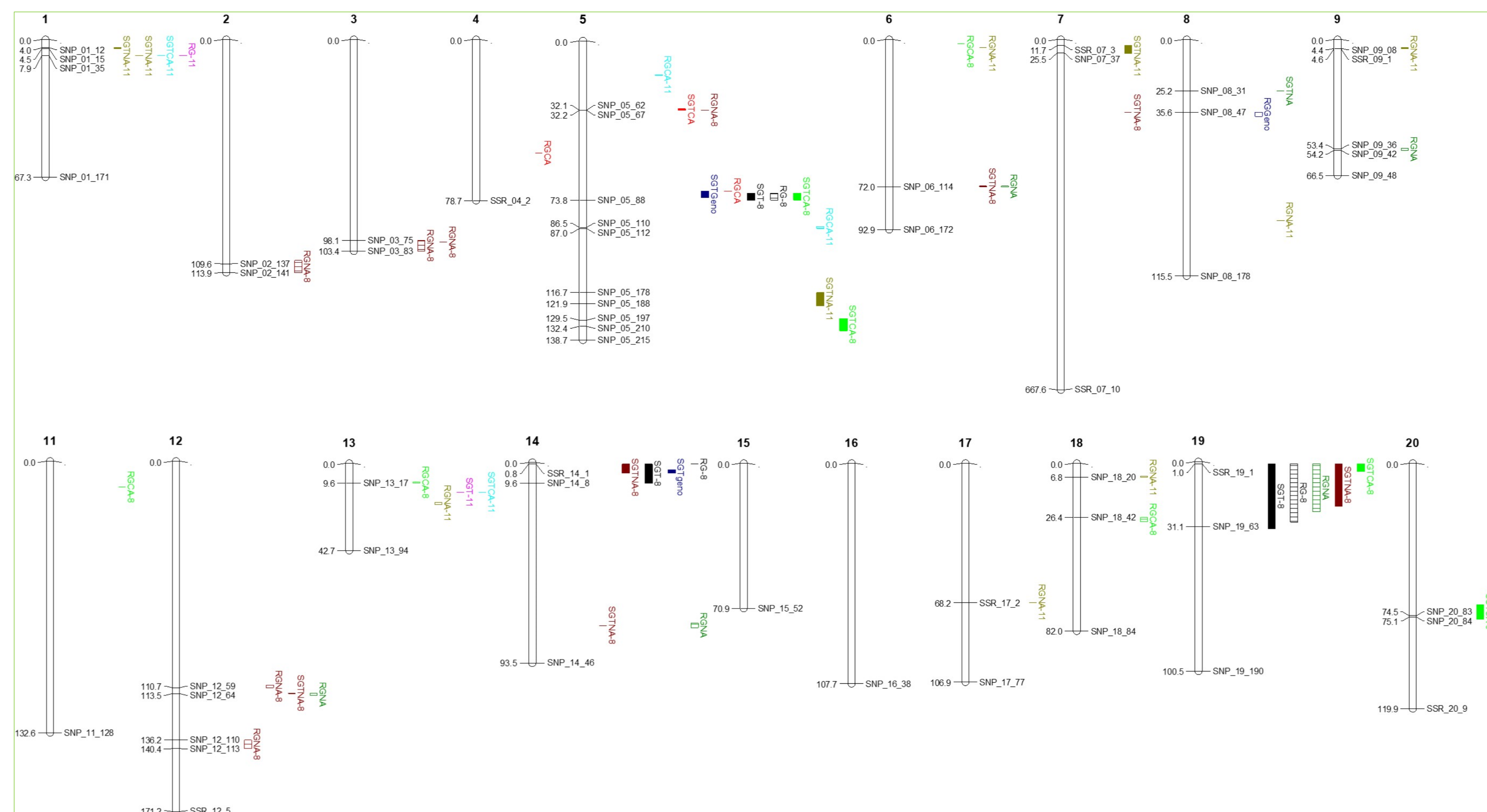
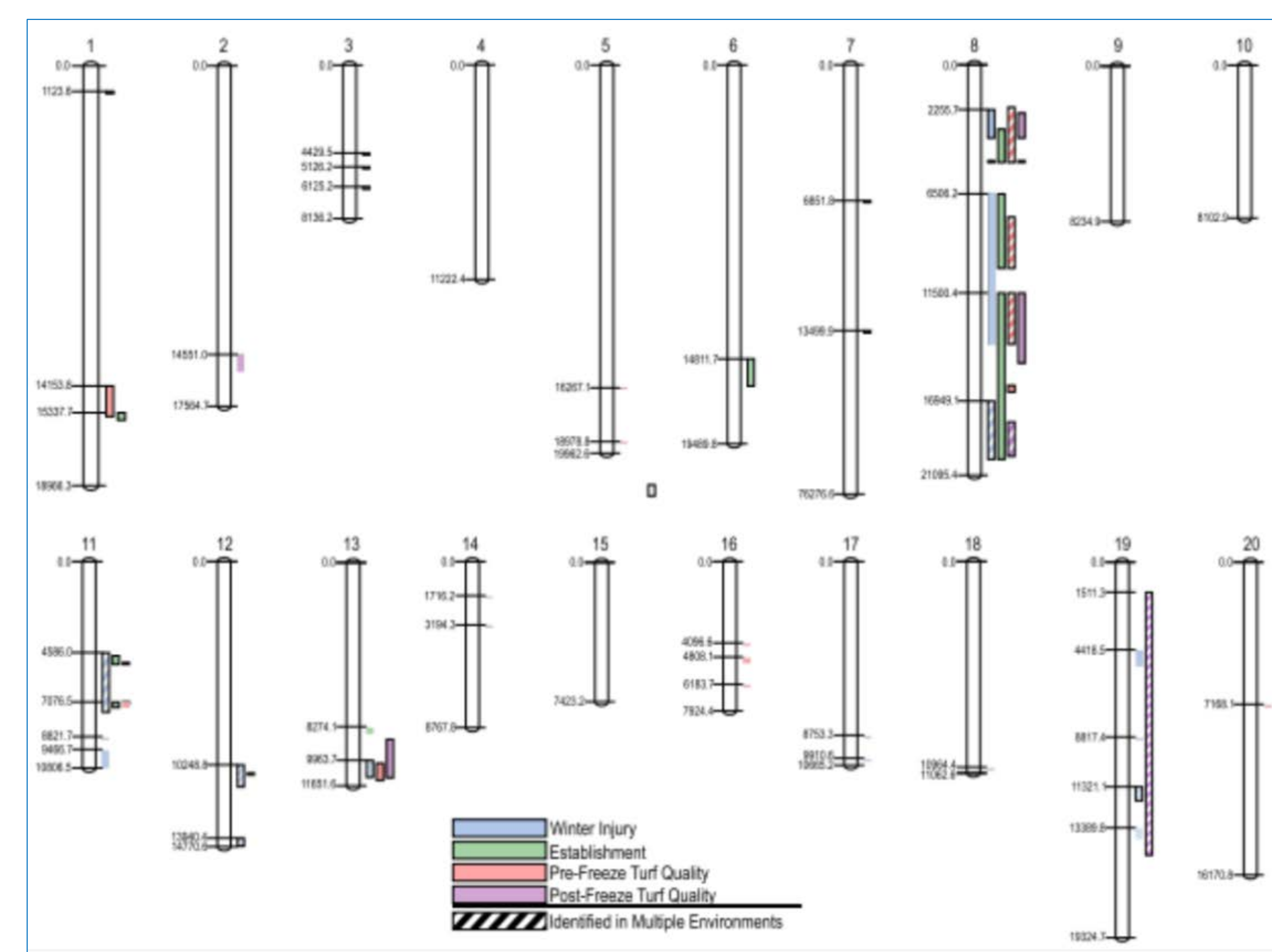
### QTL Mapping

- Analyze the freeze data with the high density linkage map (Holloway et al., 2018)
- Identify putative regions of interest by way of LS means from freeze data
- Screened for cofactors and established LOD threshold
- MQM mapping → narrow down regions of interest
- Assess co-localization of winter survival QTL (Holloway et al. 2018) with acclimation and freeze-tolerance QTL.



## QTL Mapping Results

- Right: Acclimation and freeze tolerance QTL
- Bottom: winter survival QTL (Holloway et al. 2018)
- QTL identified for multiple traits and environments.
- Regions of interest for acclimation/freeze tolerance on chromosomes 5, 12, 14 and 19.
- Regions of interest for winter survival on chromosomes 8, 11, and 13
- Overlap between the two studies occurs primarily on chromosomes 8, 12, and 19.



## Proteomic Analysis (Brown et al. doi 10.1101/581488v1)

- Study of Meyer and Victoria tissues before and after cold acclimation identified several proteins of interest for cold acclimation response:
  - LEA 3, MAPK, SOD, GAST1, Phytochrome A, ATP synthase, AGP, PLD, PSII
  - ↑ starch metabolizing proteins in Meyer, ↓ photosynthesis related proteins in Victoria
- Need to determine if these protein coding regions overlap with QTL of interest.

## Conclusions

- Overlap between the QTL of interest in the two studies occurs primarily on chromosomes 8, 12, and 19.
- Overlapping regions may be of interest for further study into their role in freezing tolerance.

## Next steps

- Further assess similarities between winter survival and acclimation/freeze tolerance QTL and proteomic data.
- Conduct transcriptomic analysis of cold acclimated Meyer and Victoria.
- Identify genes/proteins at major effect regions for future breeding efforts.