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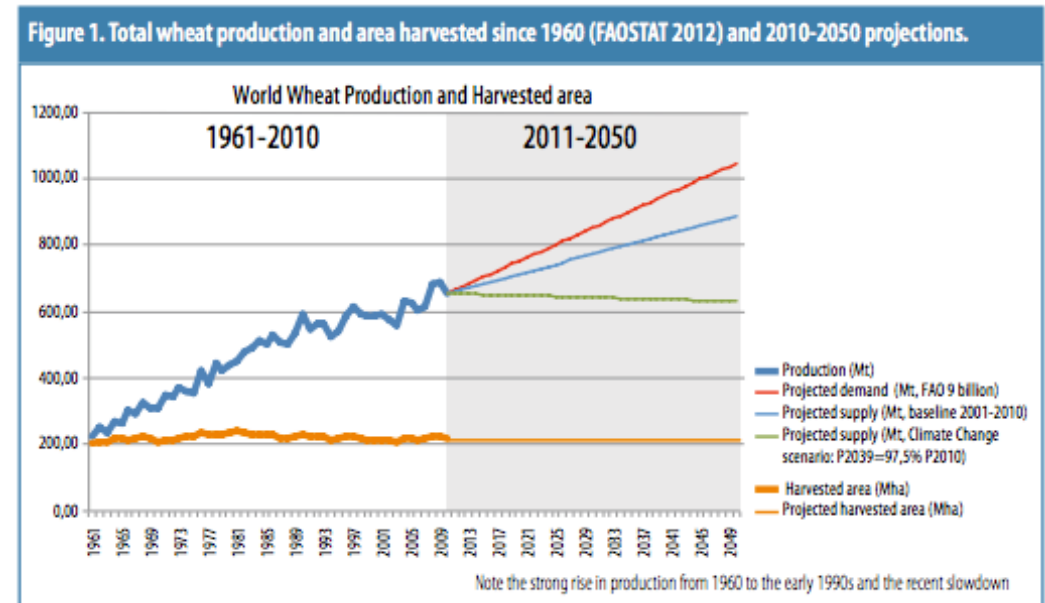
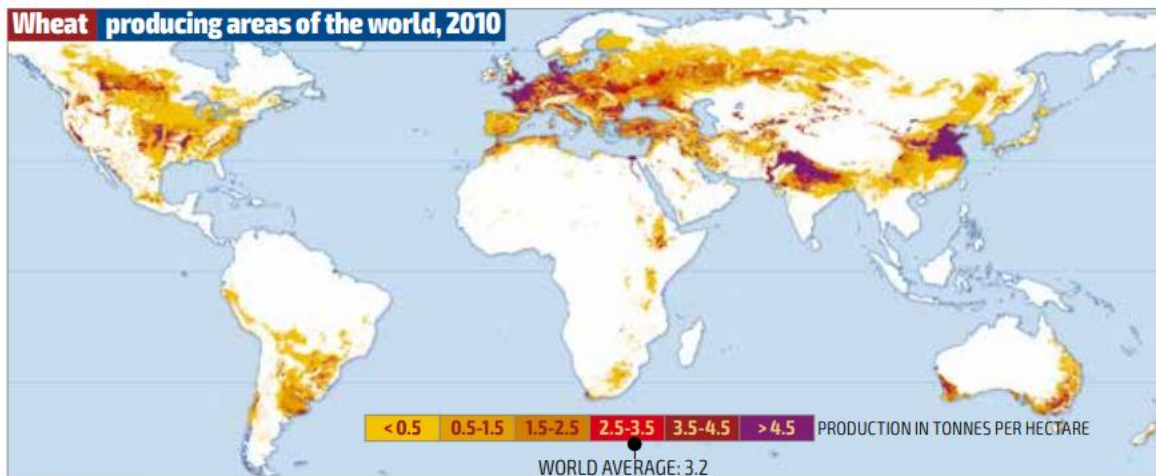
# VRNturing into the unknown: Dissecting the role of *VRN2* in climate adaptation

Dominique Hirsz

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# Climate change

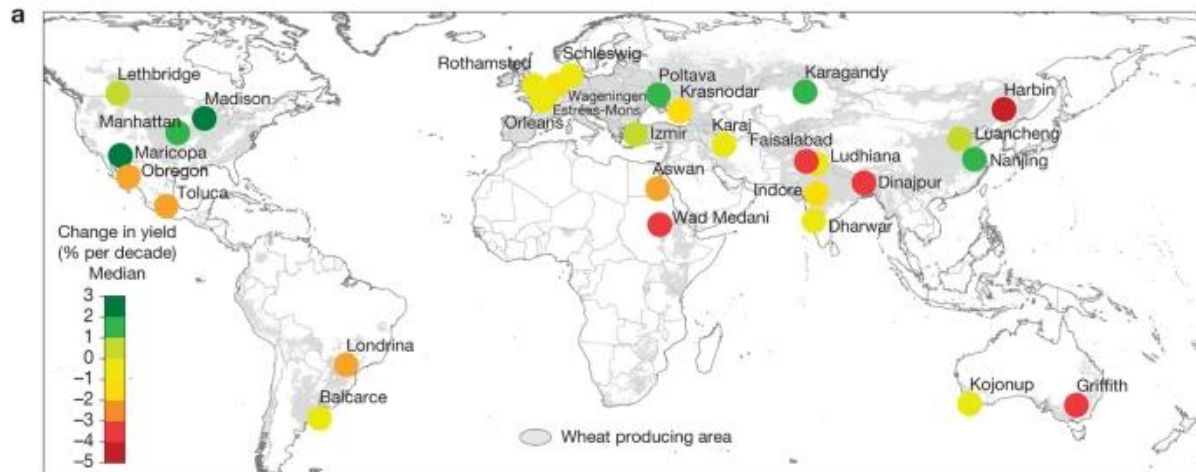
- Wheat is the most **widely** cultivated cereal crop due to **well-adapted** varieties:
  - Spring** wheat vs **winter** wheat
  - Photoperiod insensitivity
- The most vulnerable cereal to the effects of climate change
  - Each 1 °C increase is expected to reduce yield by 4-20% depending on the region



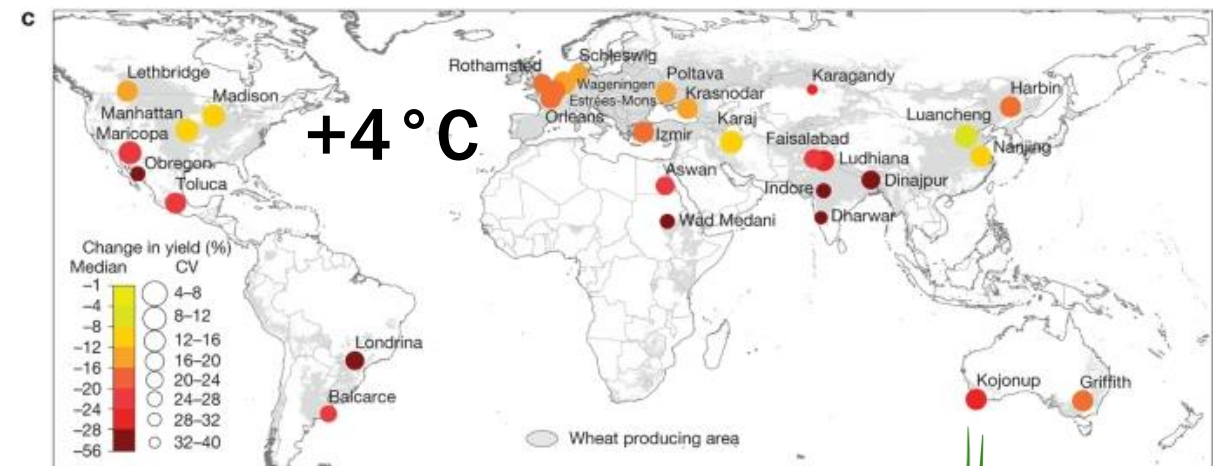
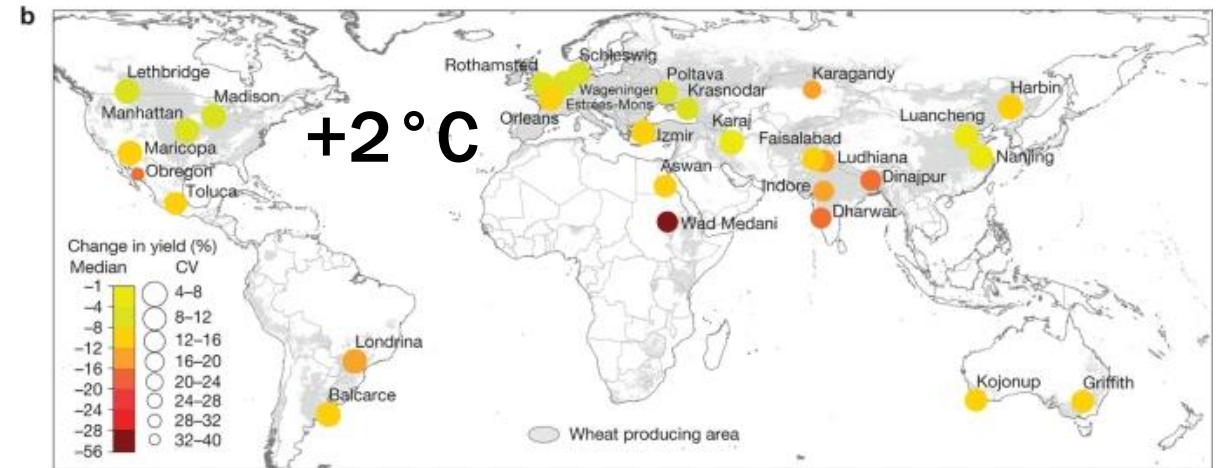
FAO, 2012

# The impacts of climate change on wheat production

## Grain trend yields from 1981 - 2010



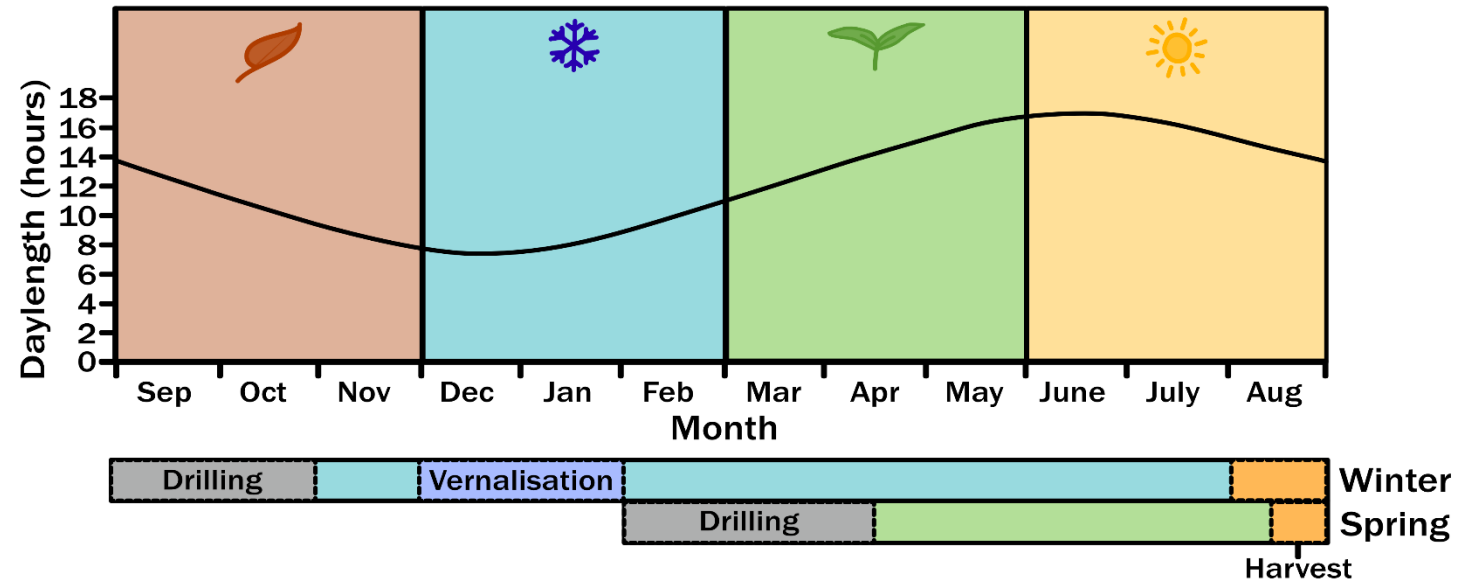
## Relative predicted grain yield for:



Asseng et al., 2015

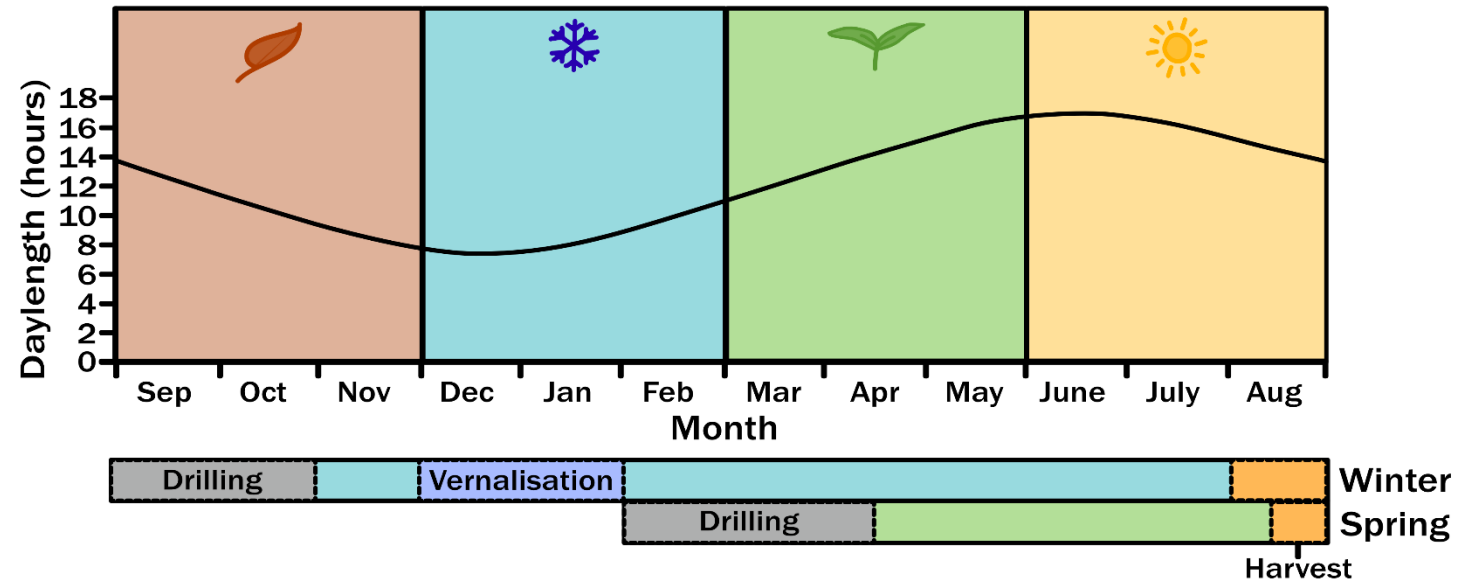
# Growth habit in cereals

- **Spring** growth – delayed drilling date, vernalisation **not required** to flower
- **Winter** growth – autumn drilling, **requires** vernalisation to flower
- **Facultative** (spring) – flexible drilling date, vernalisation **responsive** but not required to flower



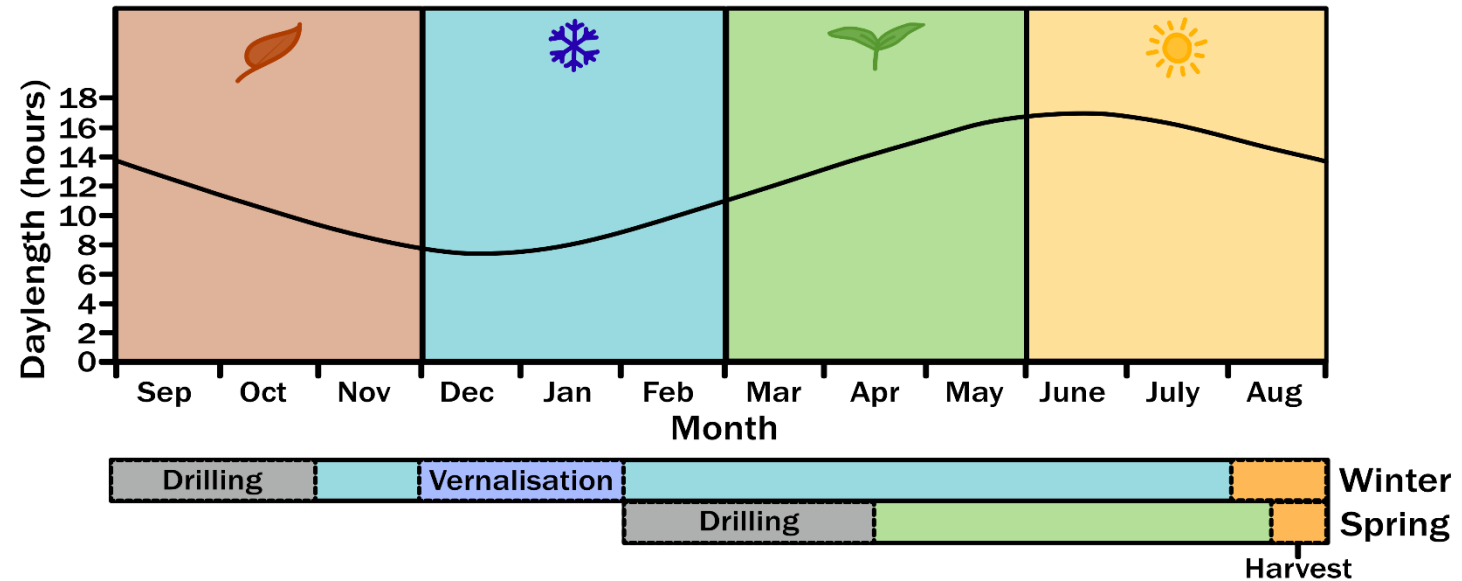
# What is vernalisation?

The requirement for an extended **cold** exposure in **winter** varieties of plants to enable **flowering**

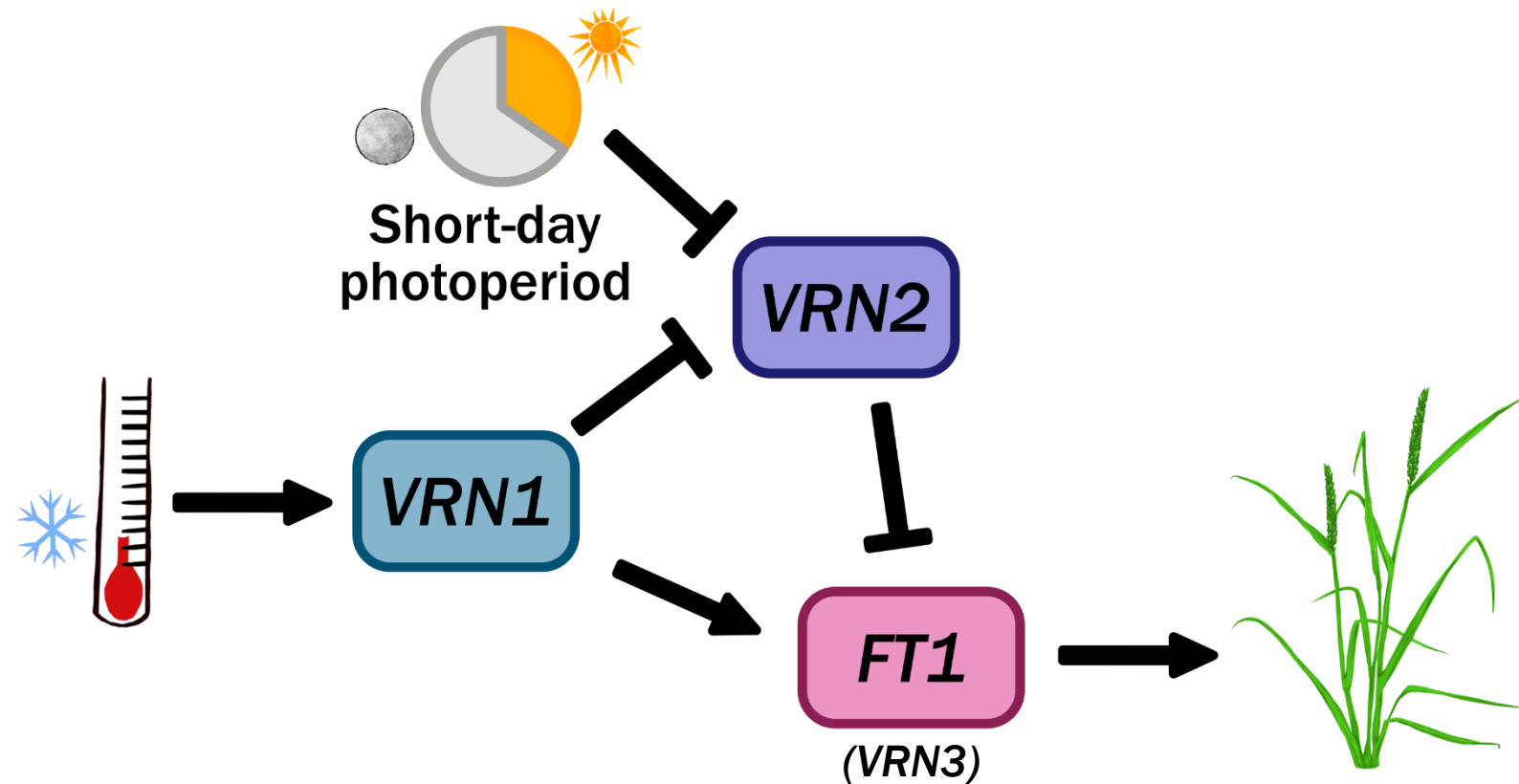


# Why work with winter wheat?

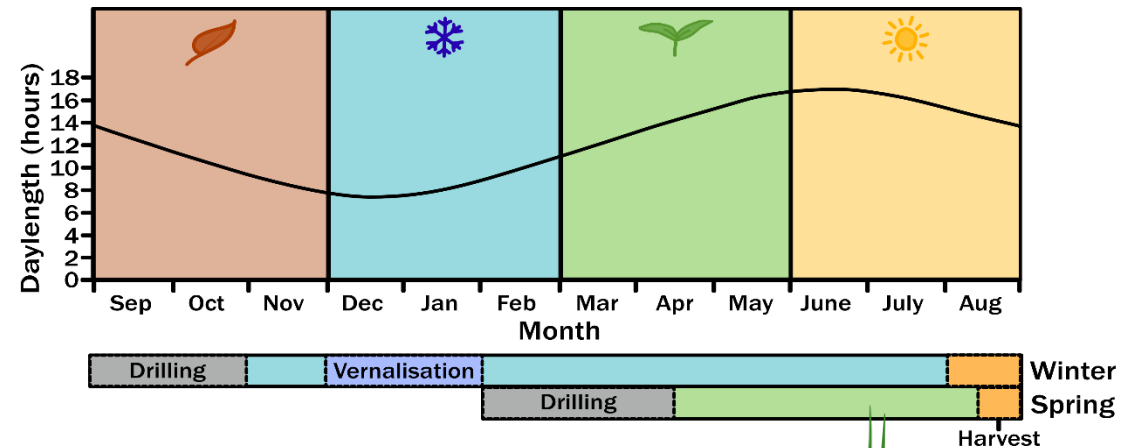
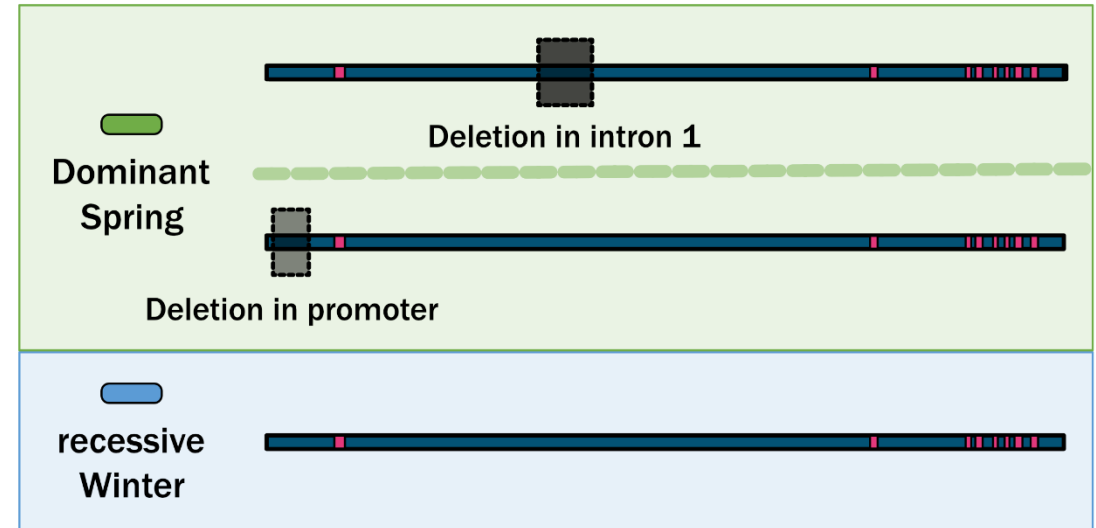
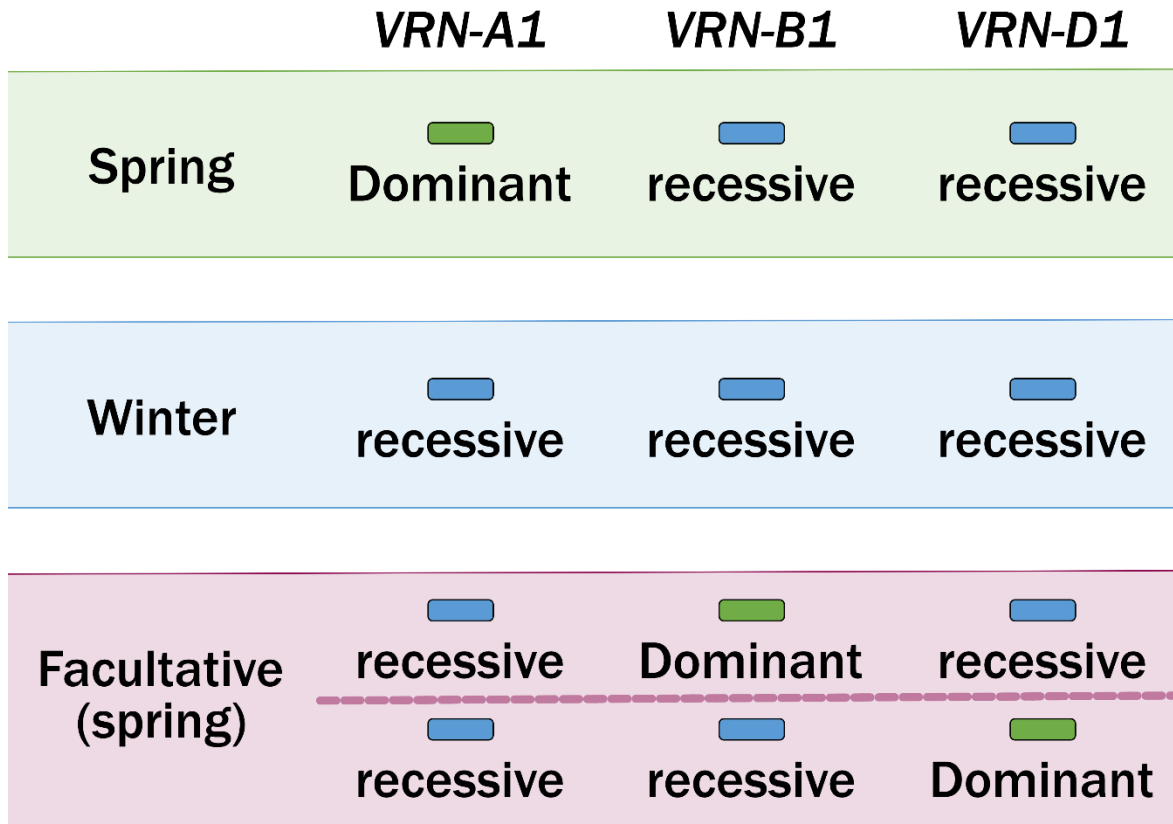
- Higher yields as flowering occurs at lower temperatures and there is more time for grain filling
- Higher protein quality than spring – necessary for higher quality flour which is essential for some baked goods (eg bread)
- Ground coverage over winter



# The vernalisation pathway in cereals



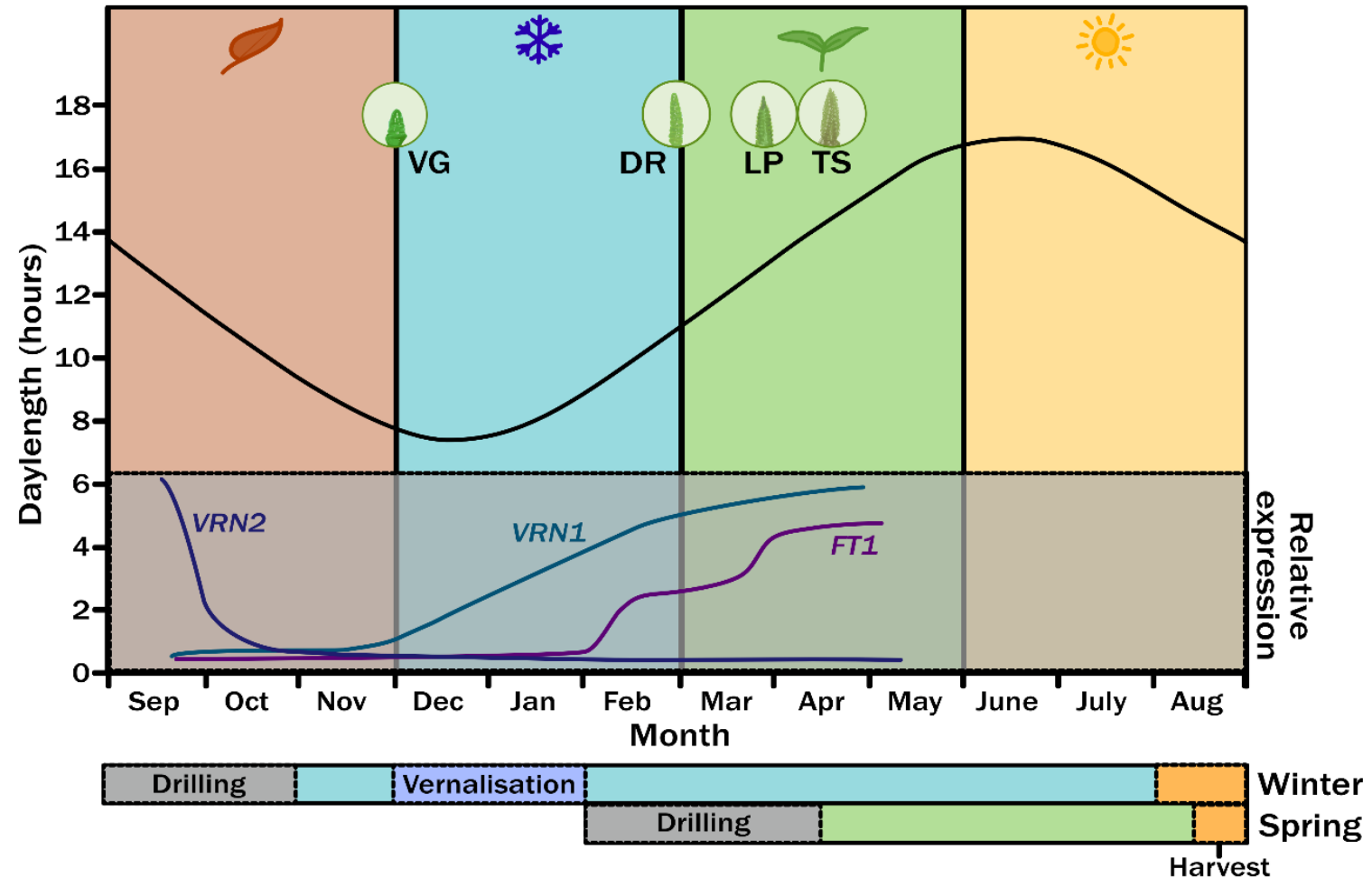
# Genetic variation underpins growth habit in cereals





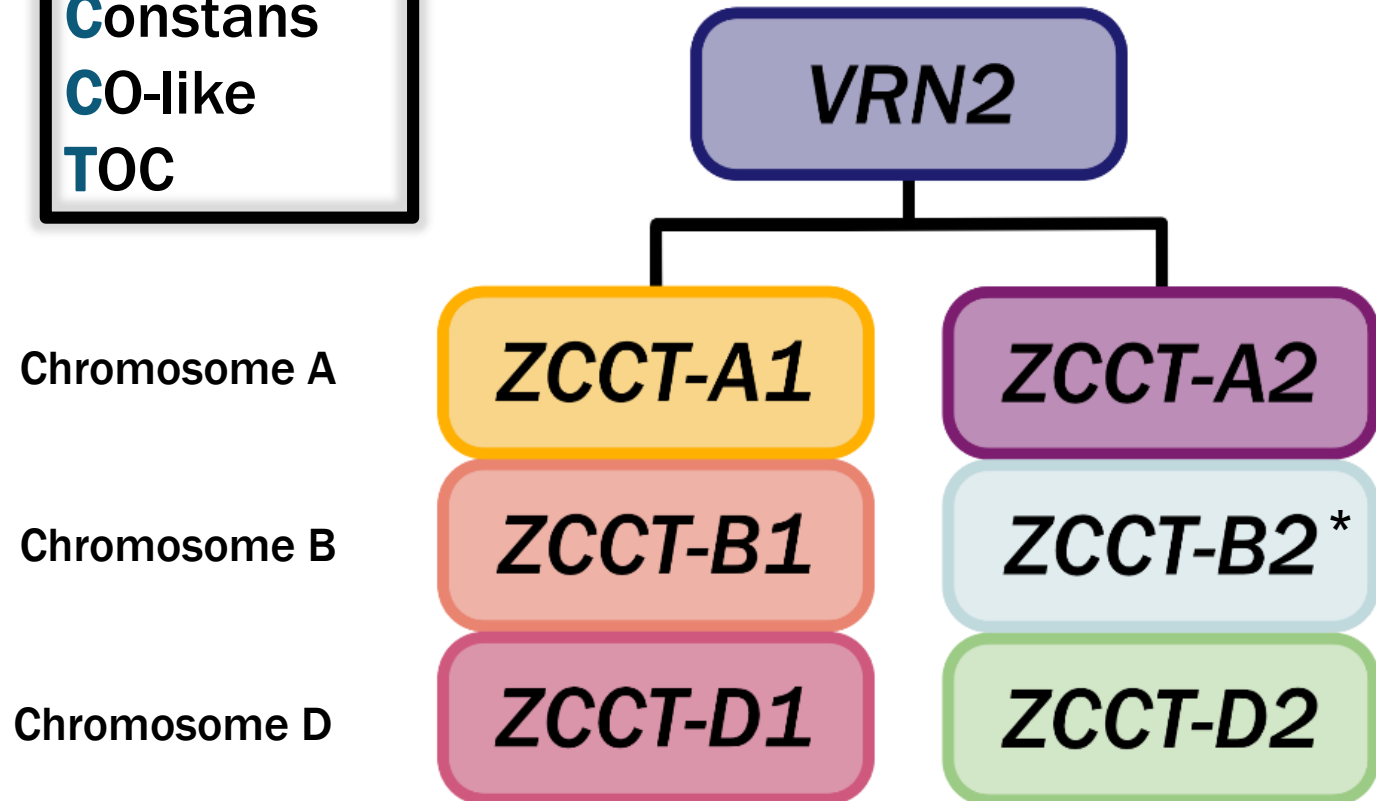
# Genetic variation underpins growth habit in cereals

- *VRN1* underpins growth habit
- *VRN2* (hexaploid) fine tunes vernalisation and provides adaptive benefit
  - Understand the effect of specific alleles

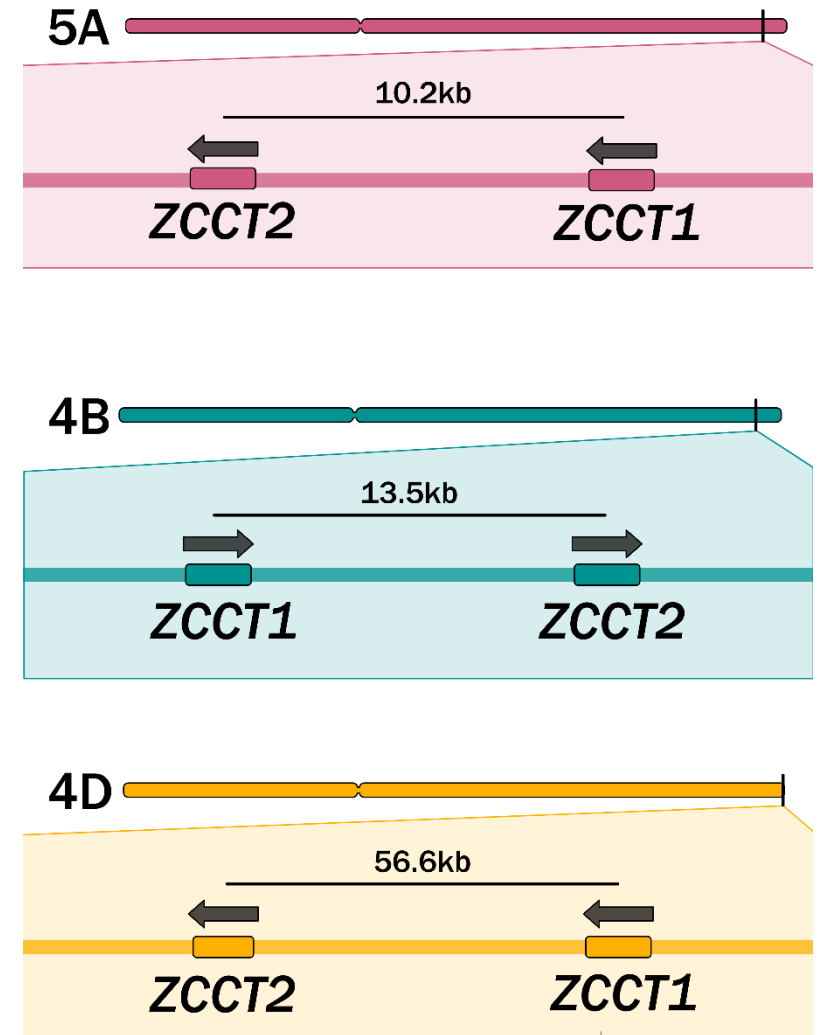


# VRN2 loci is formed of ZCCT1 and ZCCT2

**Z**inc finger  
**C**onstans  
**CO**-like  
**T**OC



\*not present in the reference sequence



# The CCT domain is well conserved

## C2H2 ZINC FINGER



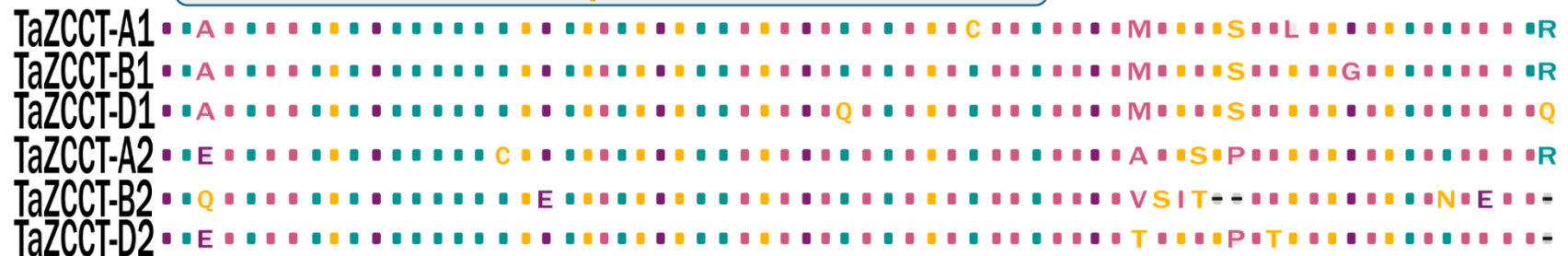
Zinc finger  
Constans  
CO-like  
TOC

## TPFHETAAAGNS.SRLTLEVSGGGRHMAHLYQPPA--RAHIVPFYGGAFNTISNEA<sup>1</sup>IMTIDTEMMVGRAHYPTMQ



## CCT DOMAIN

## ERA<sup>1</sup>AKVMRYREKRRRRYDKQIRYESRKAYAE<sup>1</sup>LRPRVNGRFVKVPEAMASPS<sup>1</sup>SPASPYDPSK<sup>1</sup>LHLGWFR<sup>1</sup>

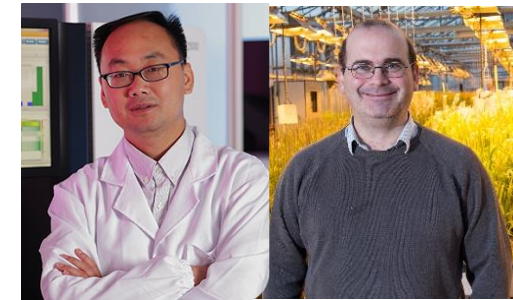
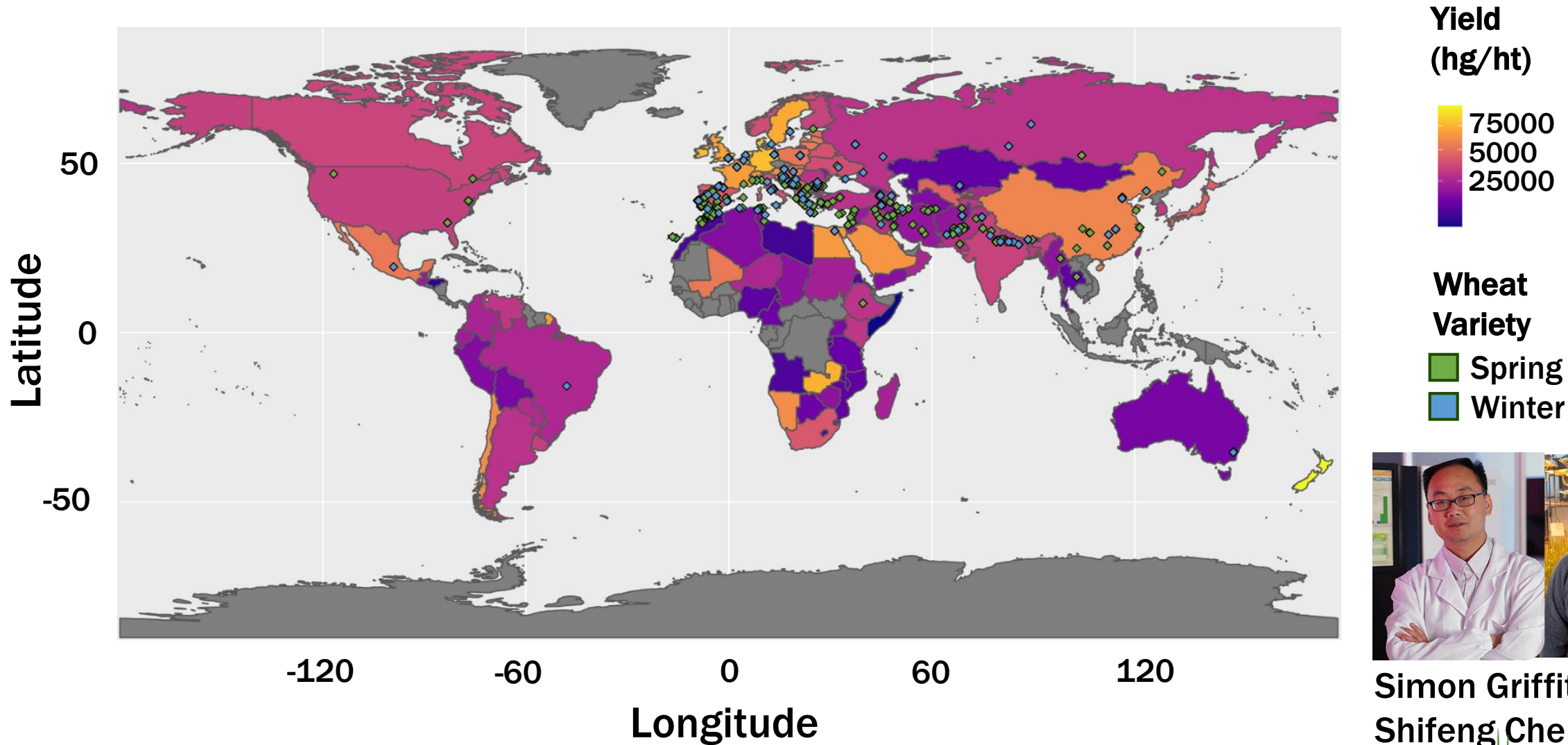


# Key Questions

 Is there an **additional** adaptive role for *VRN2*?

 Are all *ZCCTs* equal?

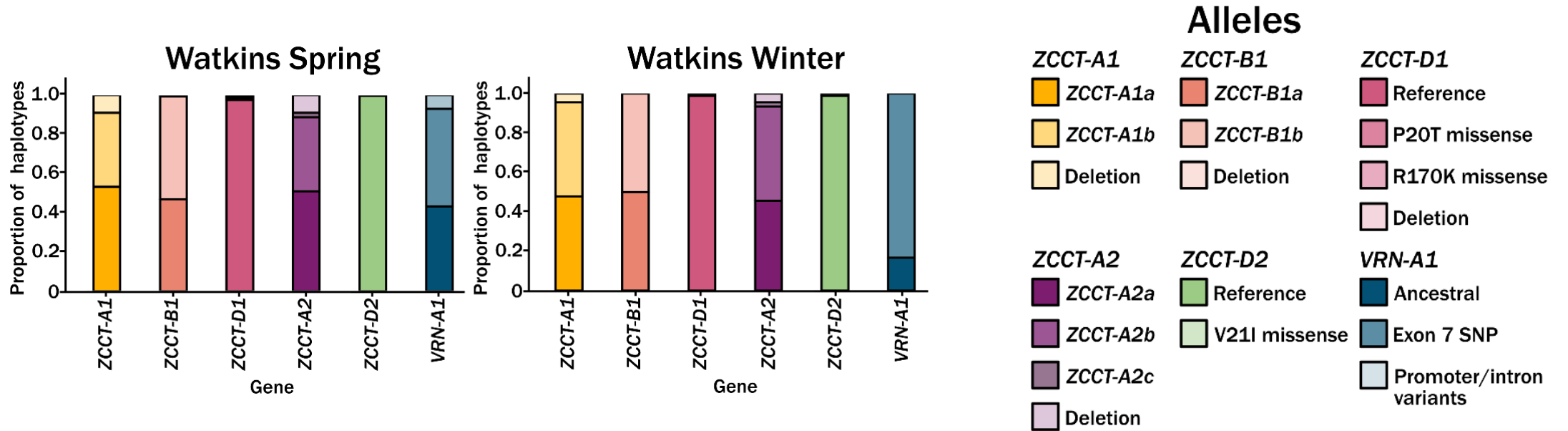
# Natural variation in the Watseq Collection



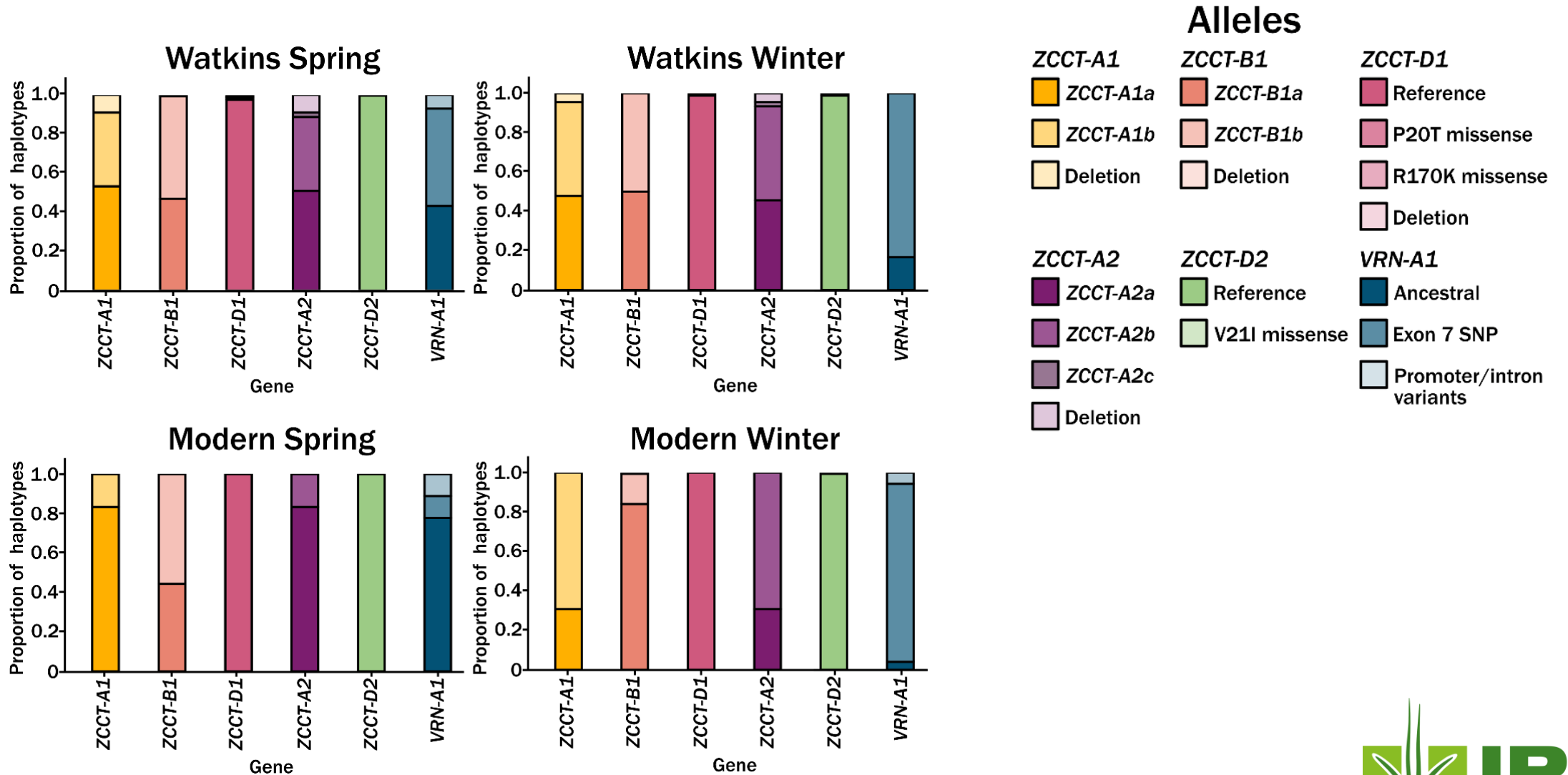
Simon Griffiths, JIC  
Shifeng Cheng, AGIS

Cheng et al., 2024

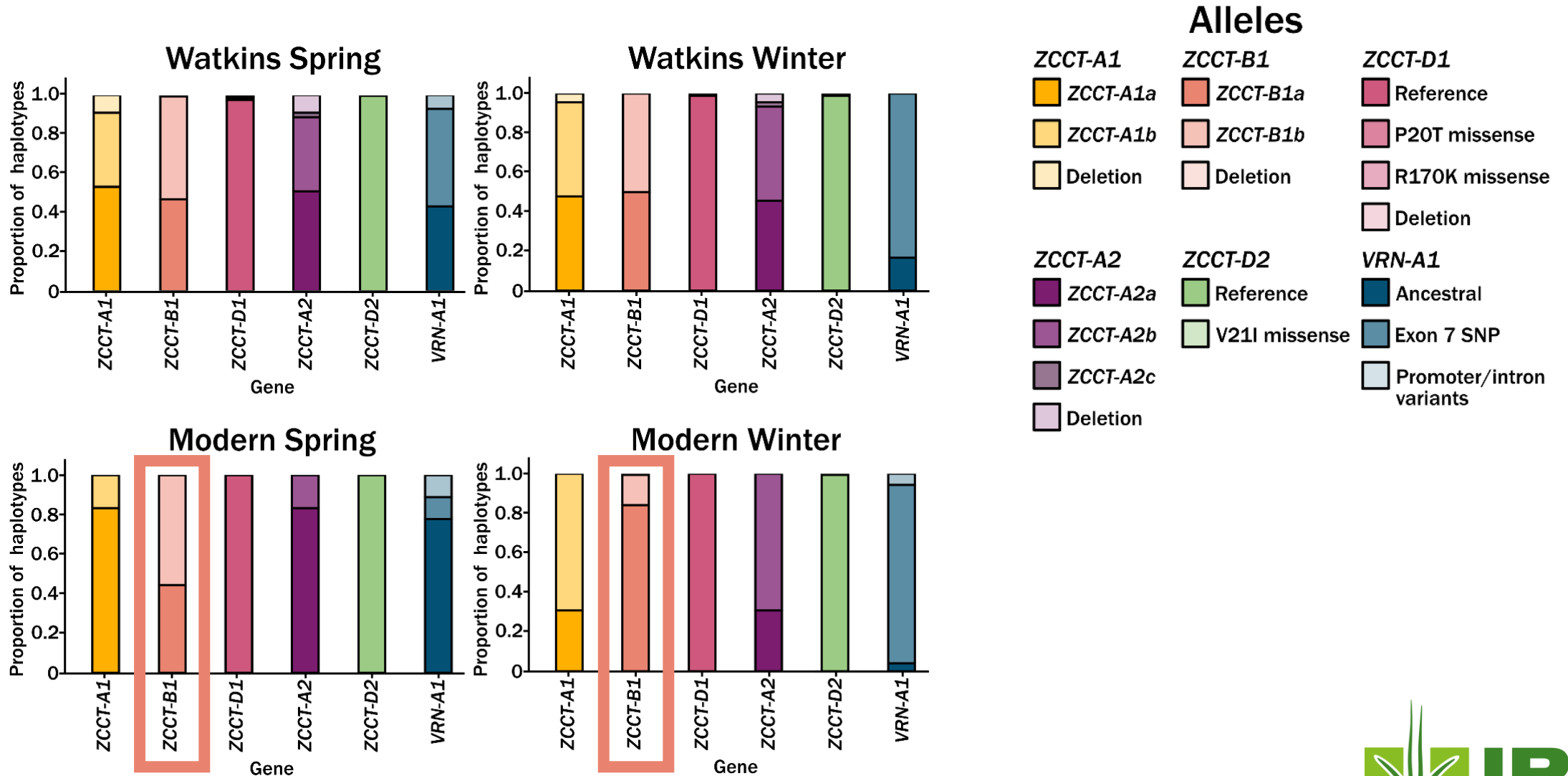
# Natural variation in the Watseq Collection



# Natural variation in the Watseq Collection

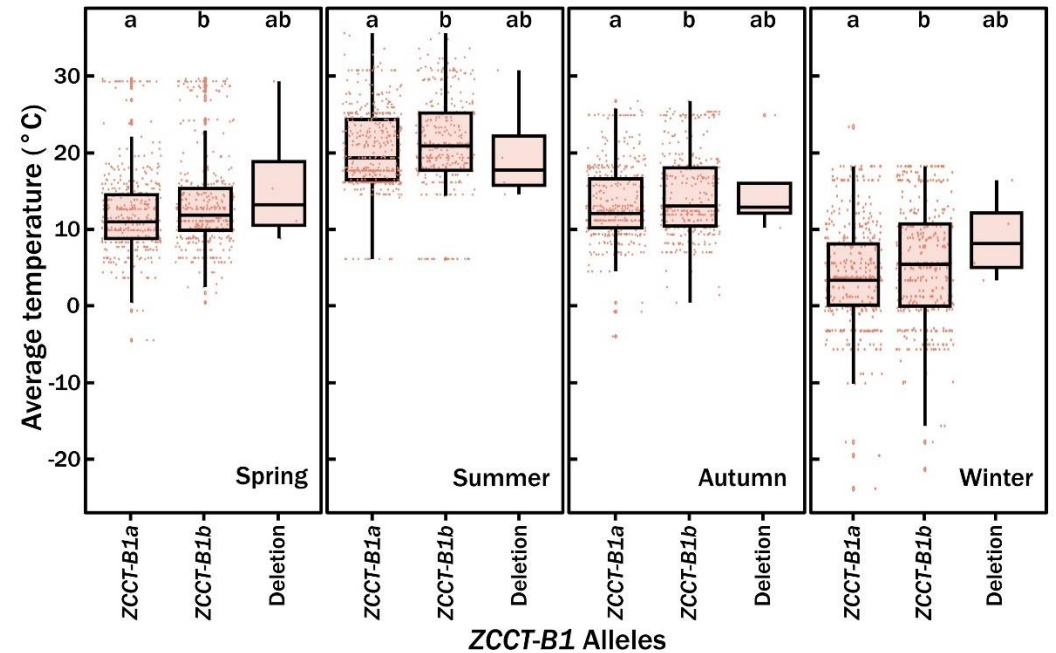
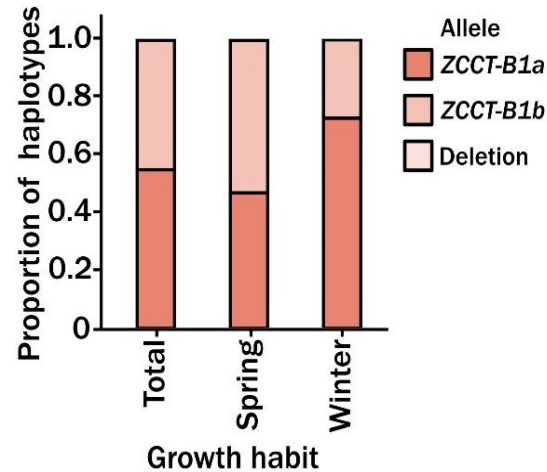
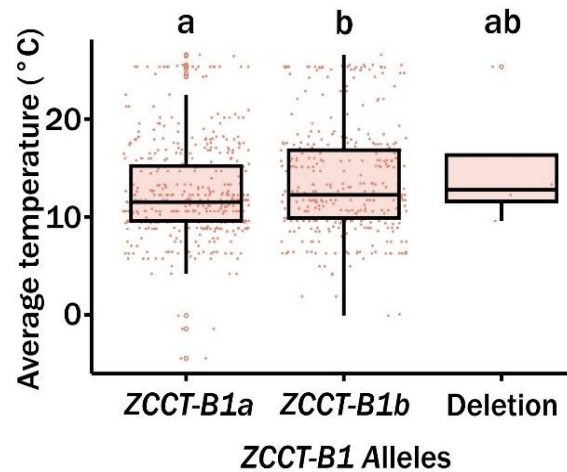


# Natural variation in the Watseq Collection

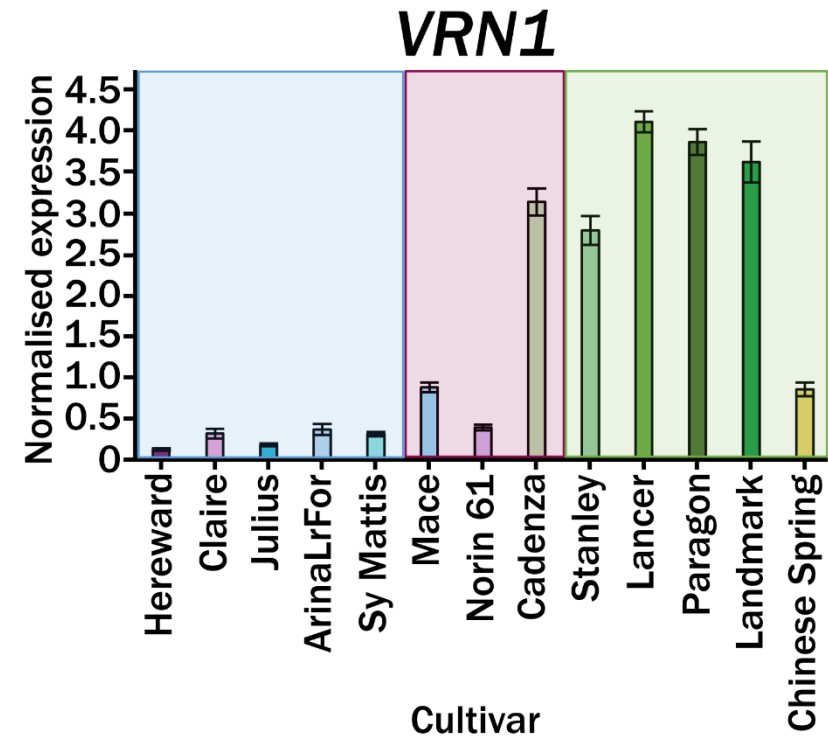
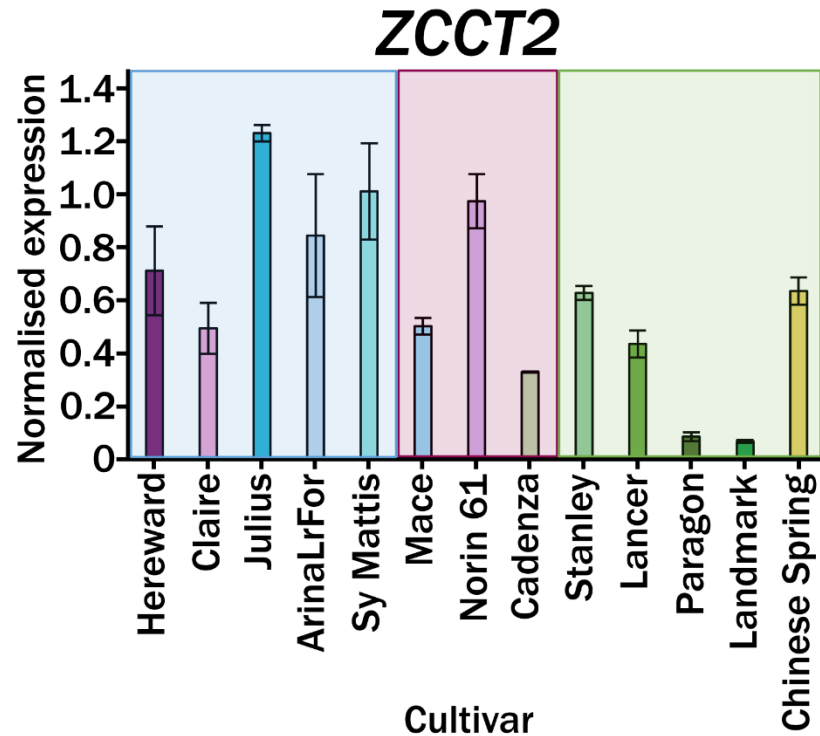
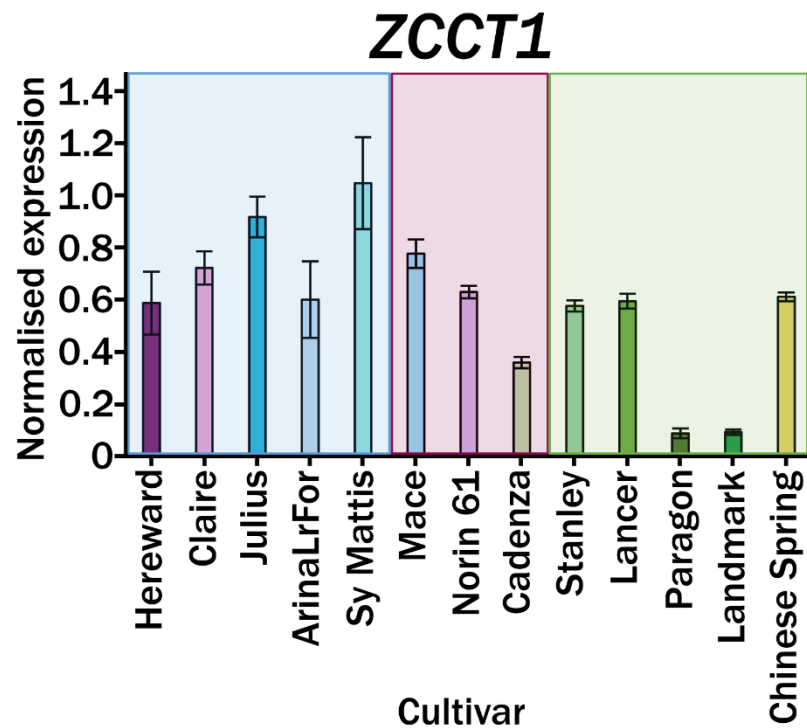




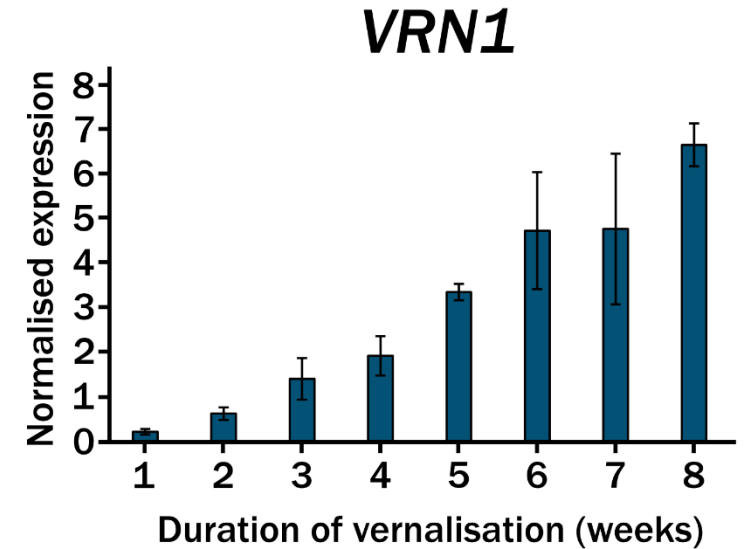
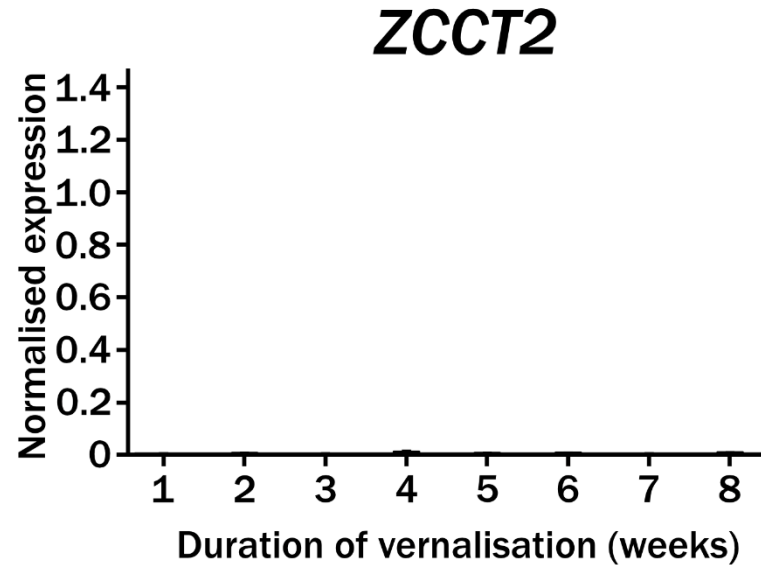
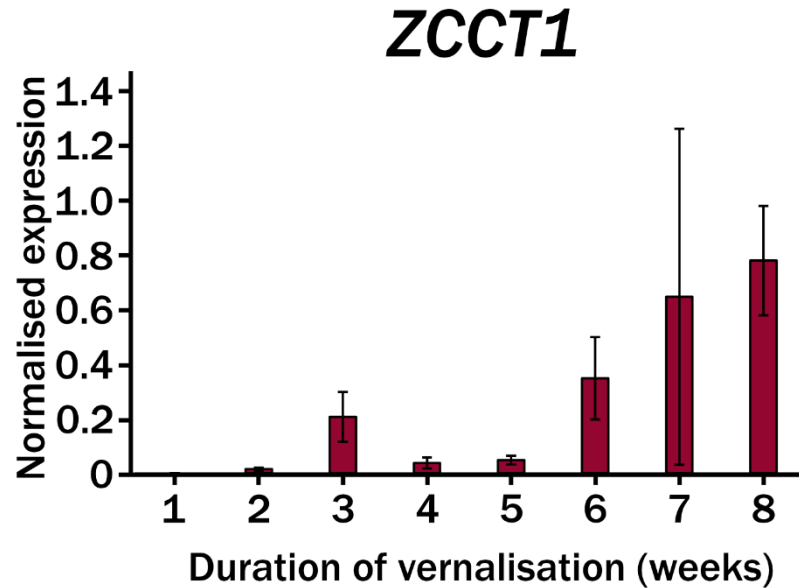
# Different alleles of *ZCCT-B1* have been selected for depending on temperature



# ZCCT expression is variable across cultivars

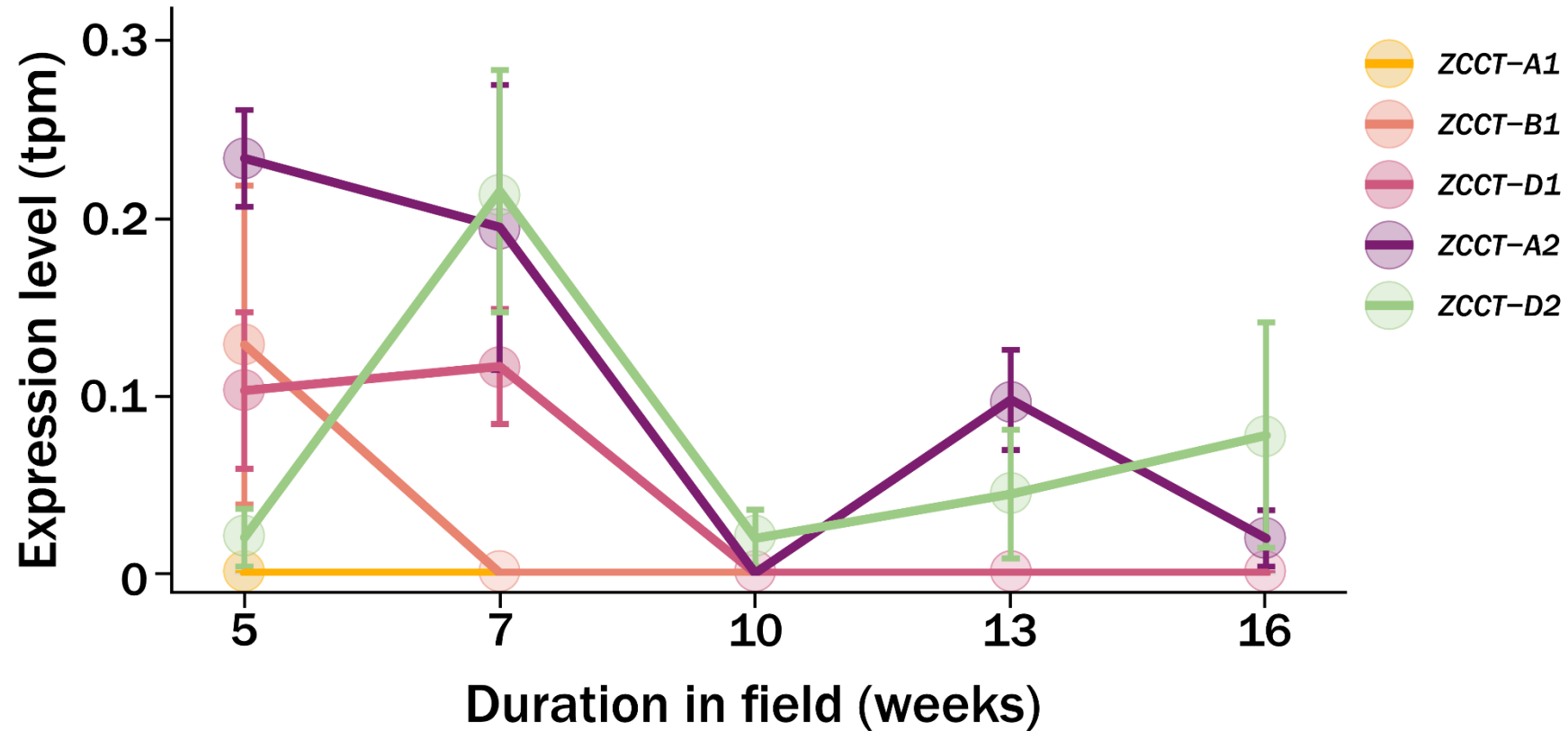


# ZCCT1 and ZCCT2 are expressed differently during vernalisation



Indi Lacey

# *VRN1* and *VRN2* expression during vernalisation in the field



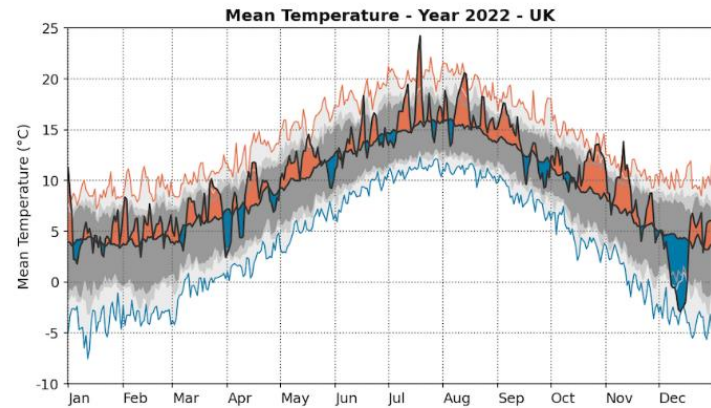
Kate O'Connor

# Condensing All Seasons INTO One (CASINO): environmental conditions

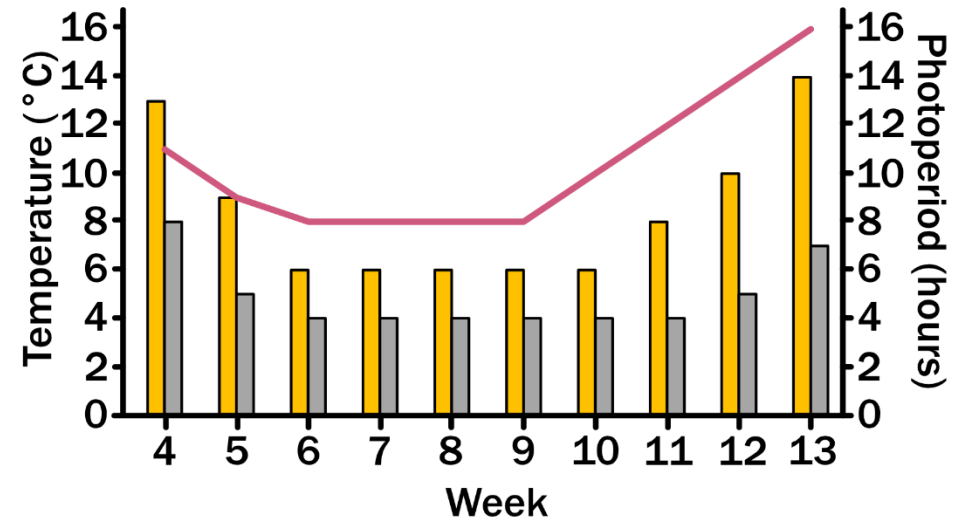
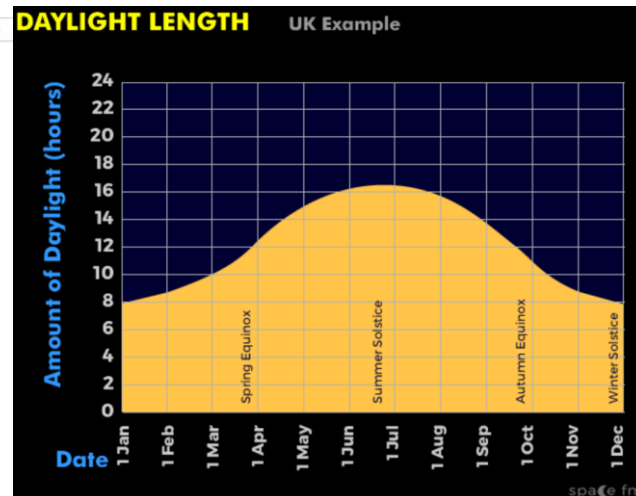
Met Office

Source: HadUK-Grid 01/01/2023 11:18

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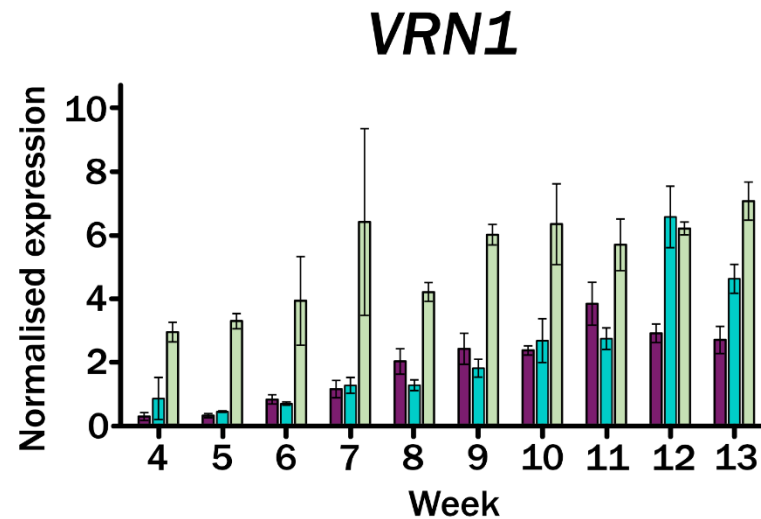
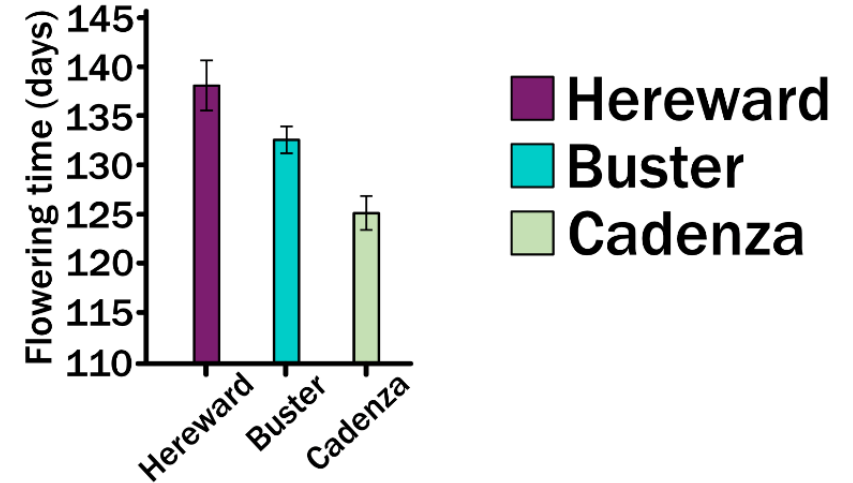
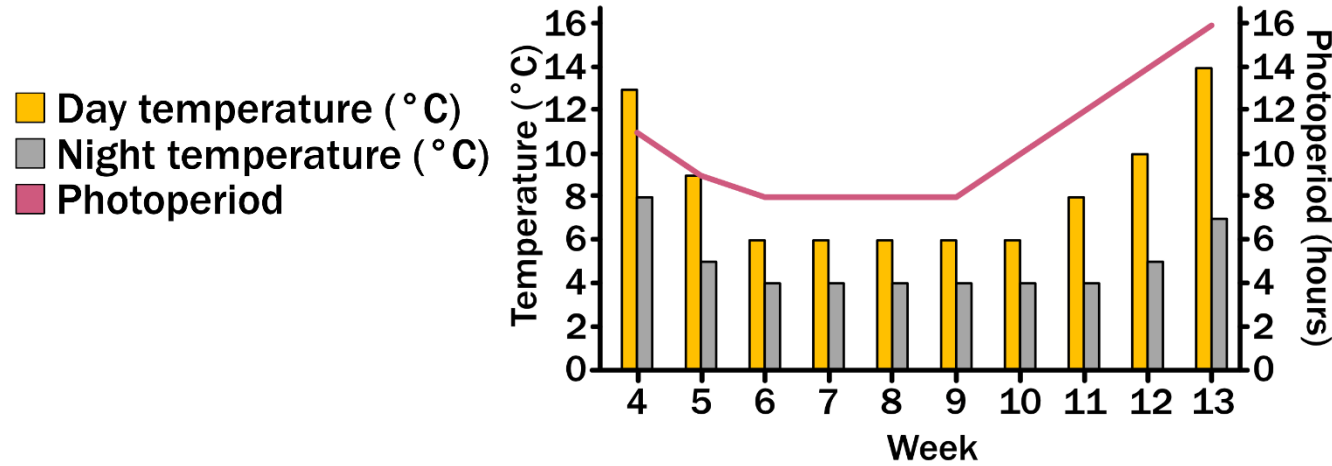


— 1991-2020 — lowest — 5% — 10% — 90%

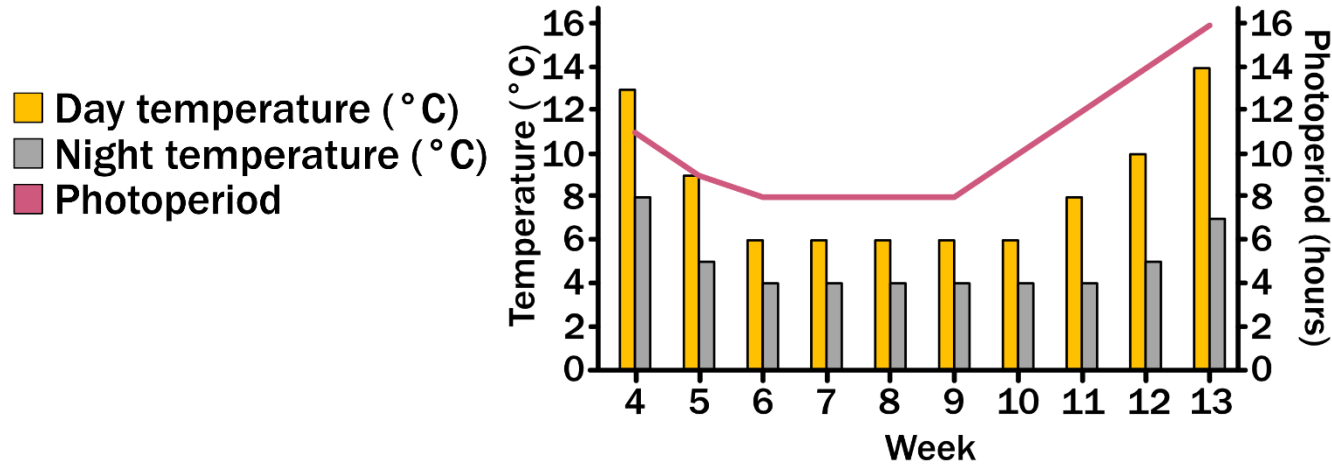


- Day temperature (°C)
- Night temperature (°C)
- Photoperiod

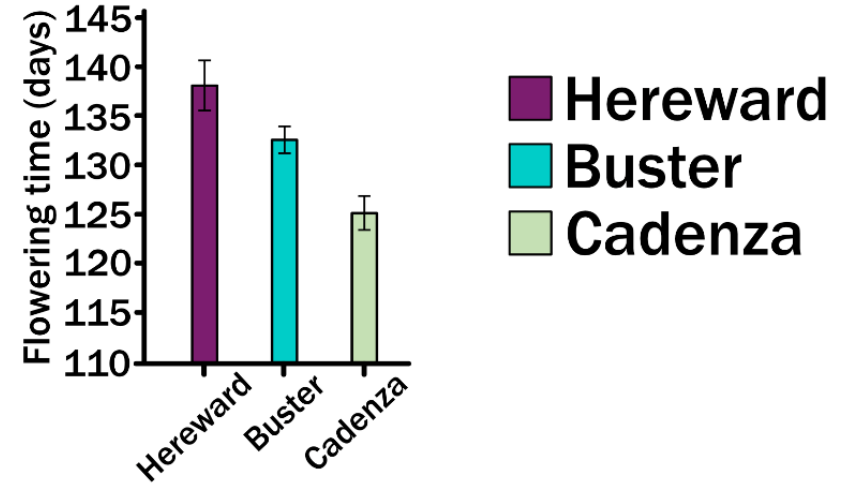
# VRN1 expression increases during vernalisation



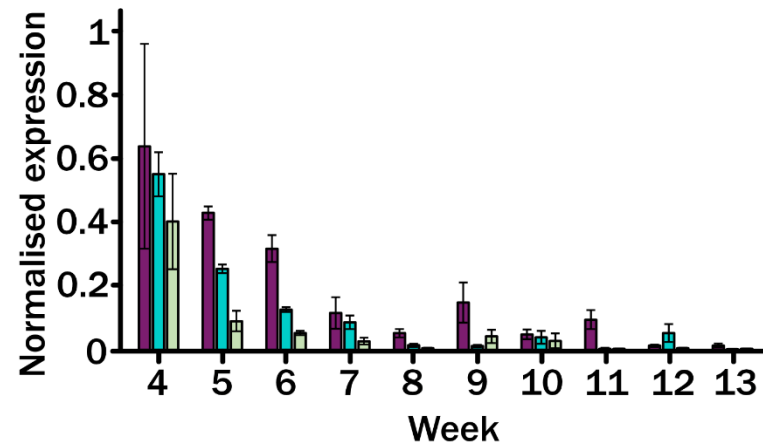
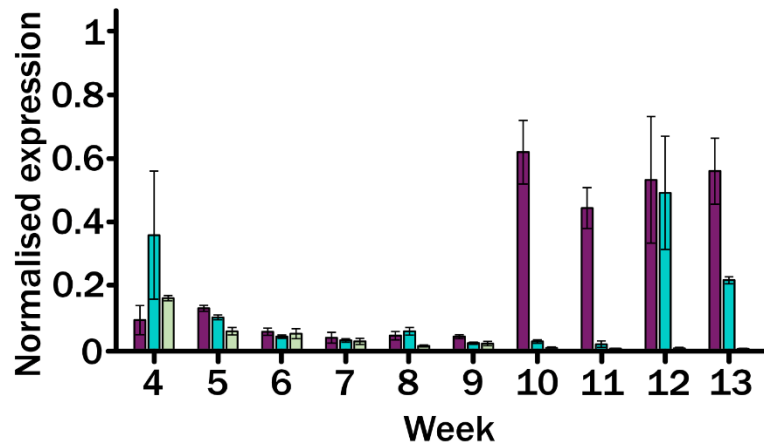
# ZCCT1 expression increases post-vernalisation



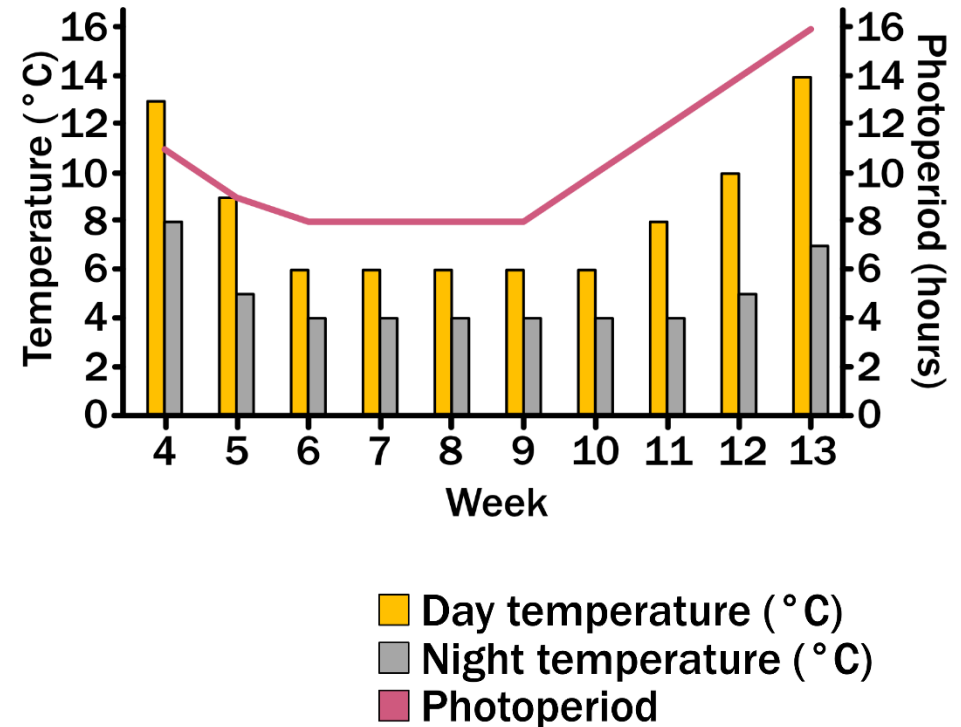
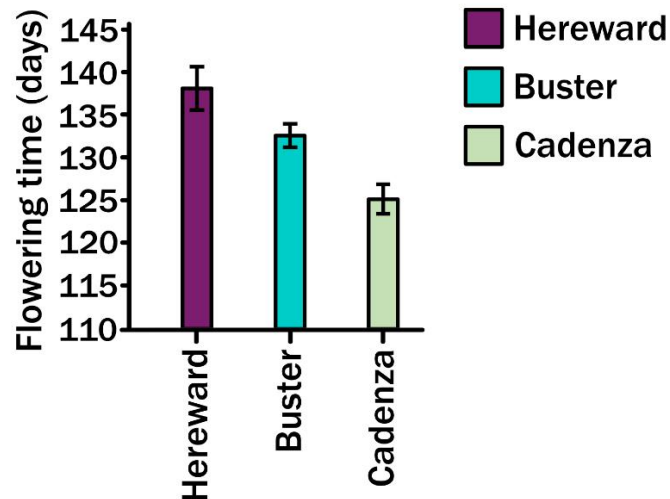
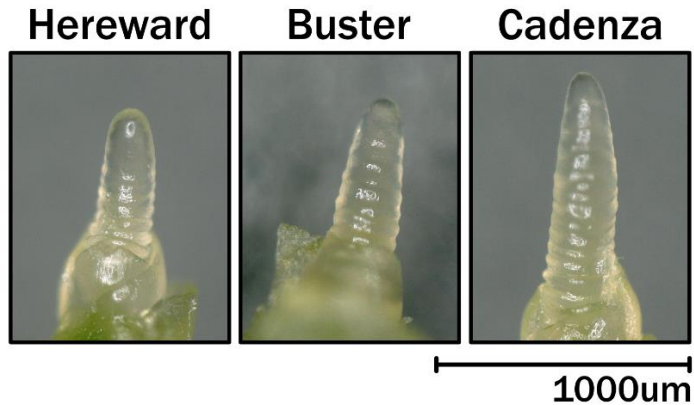
**ZCCT1**



**ZCCT2**

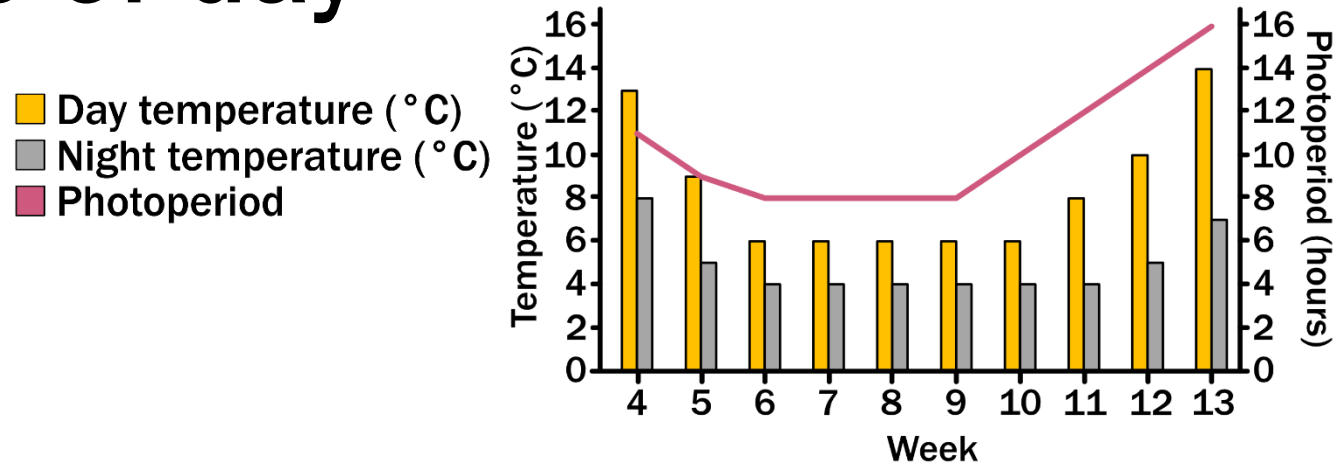


# All cultivars flowered despite the late expression of *ZCCT1*

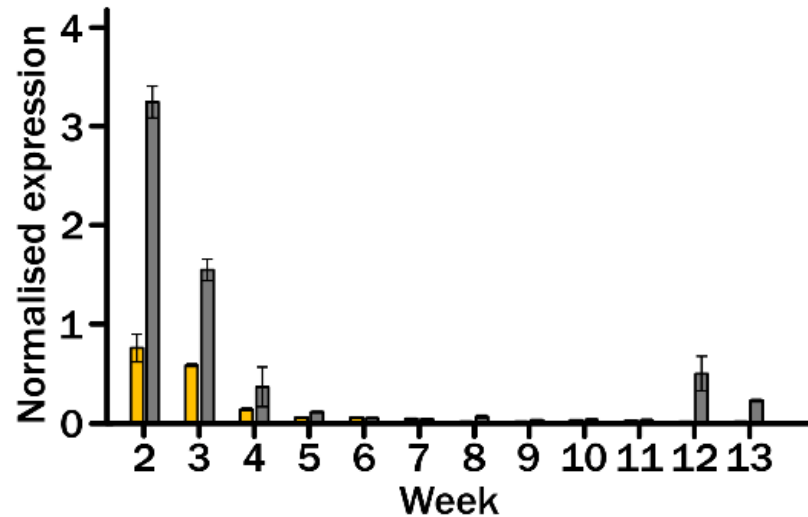




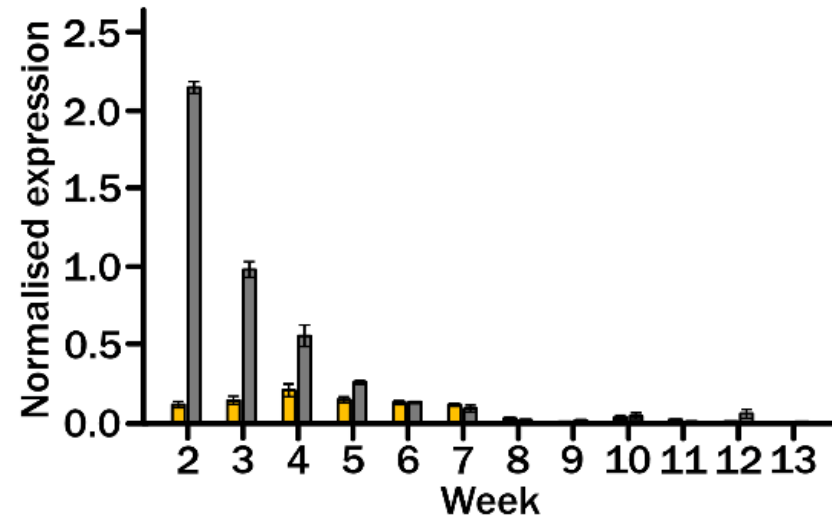
# ZCCT expression patterns vary depending on time of day



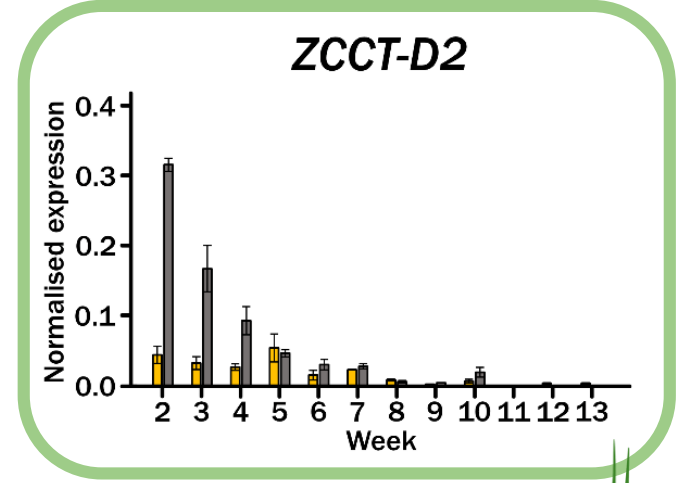
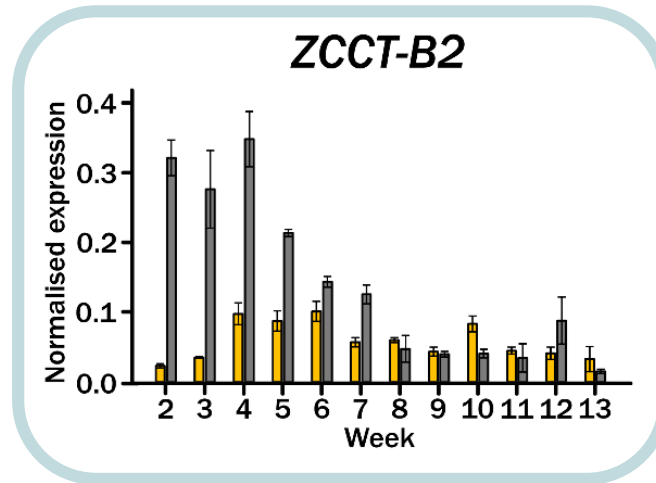
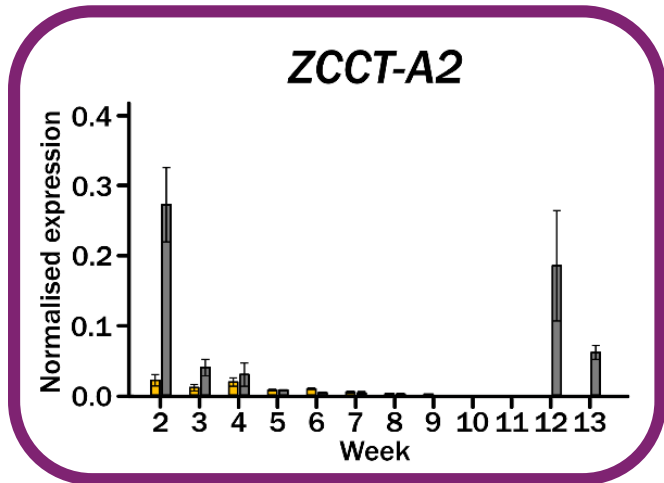
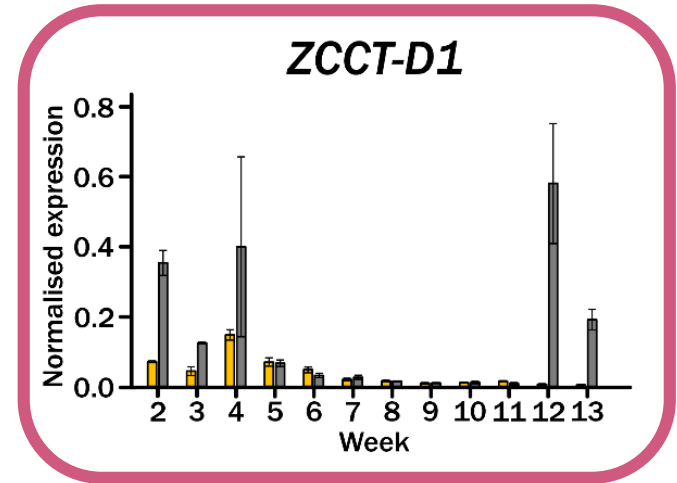
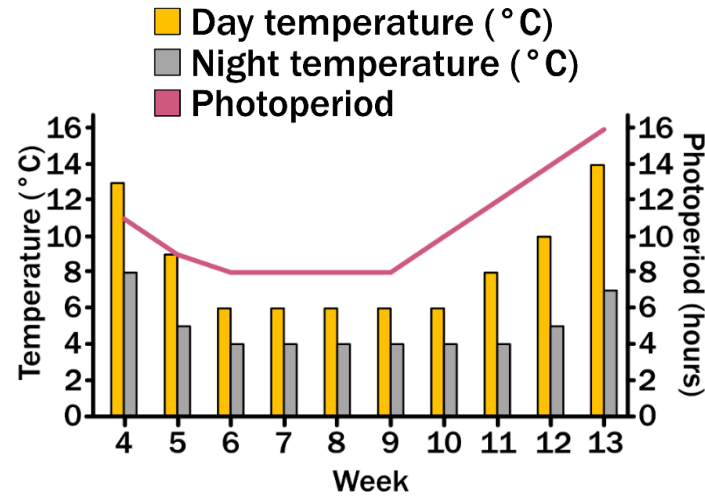
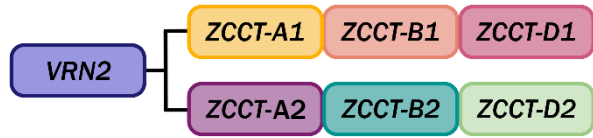
## ZCCT1



## ZCCT2

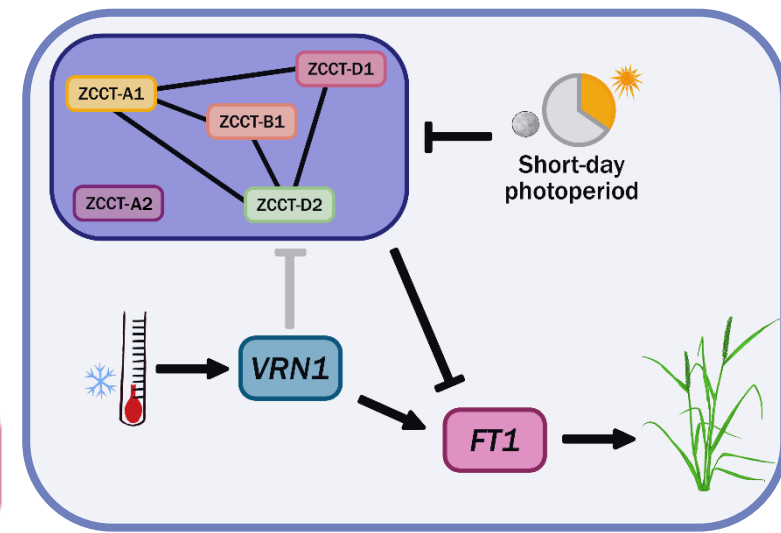
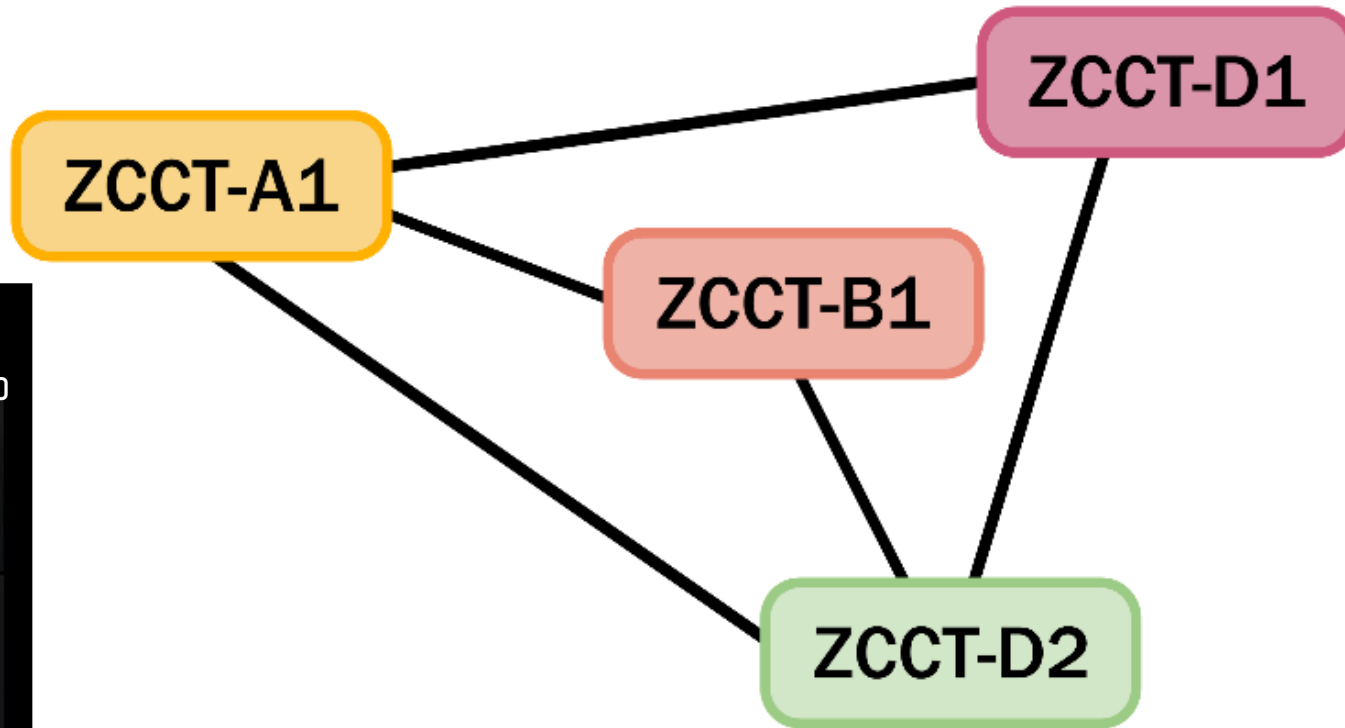


# Different *ZCCT* genes show varying expression patterns

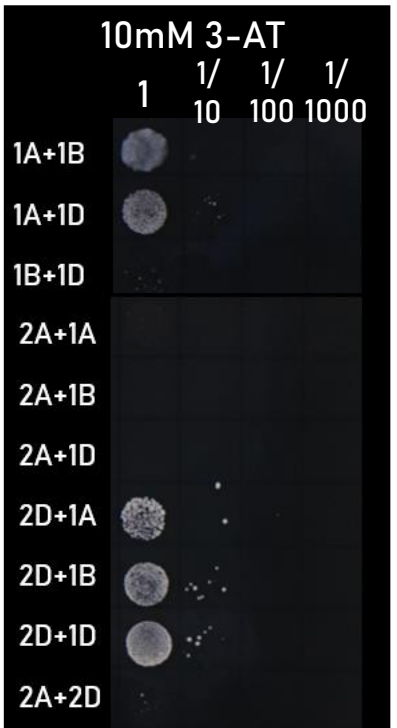


■ Dawn timepoint  
 ■ Dusk timepoint

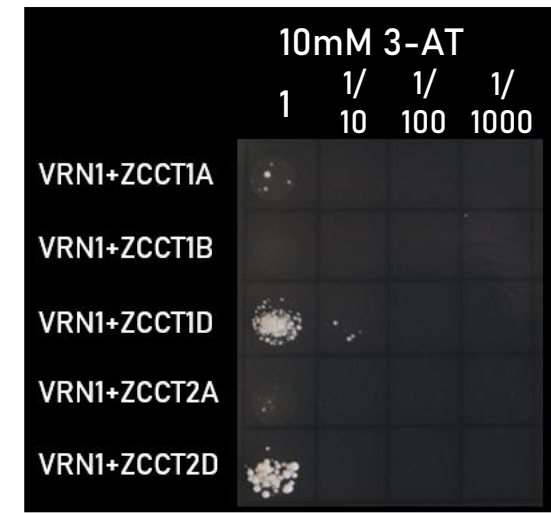
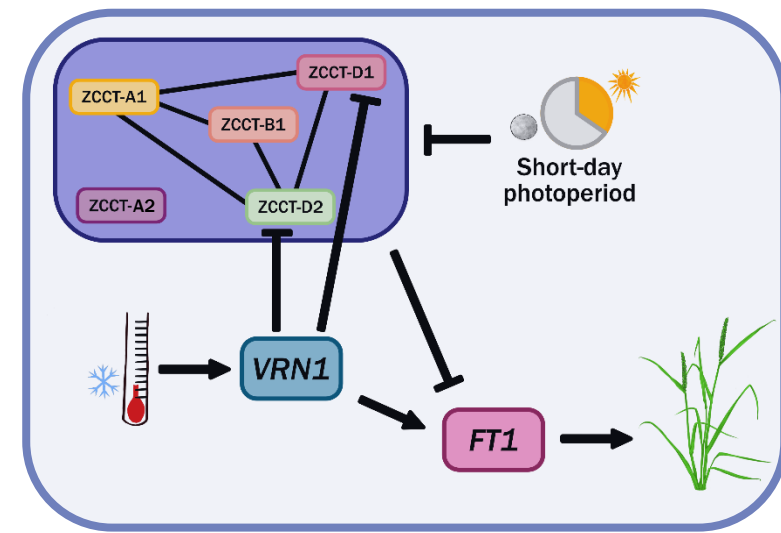
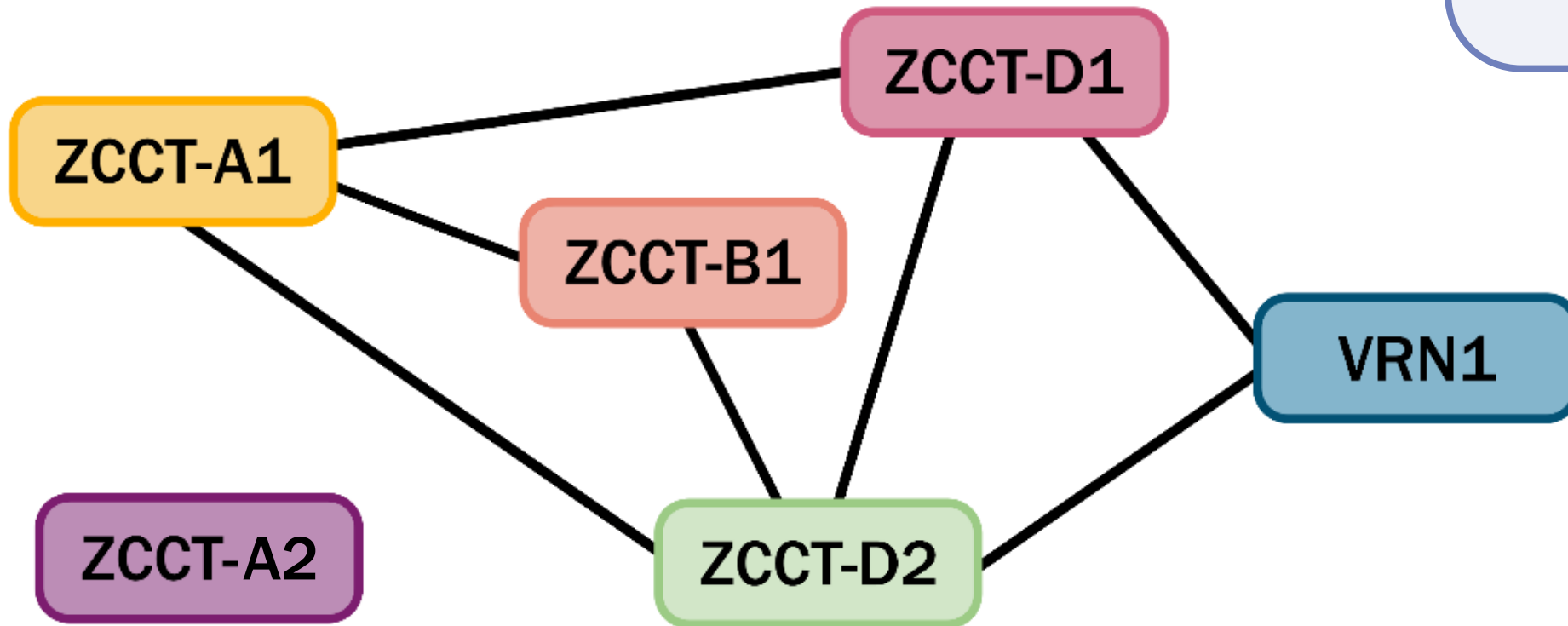
# ZCCT proteins form different heterodimers



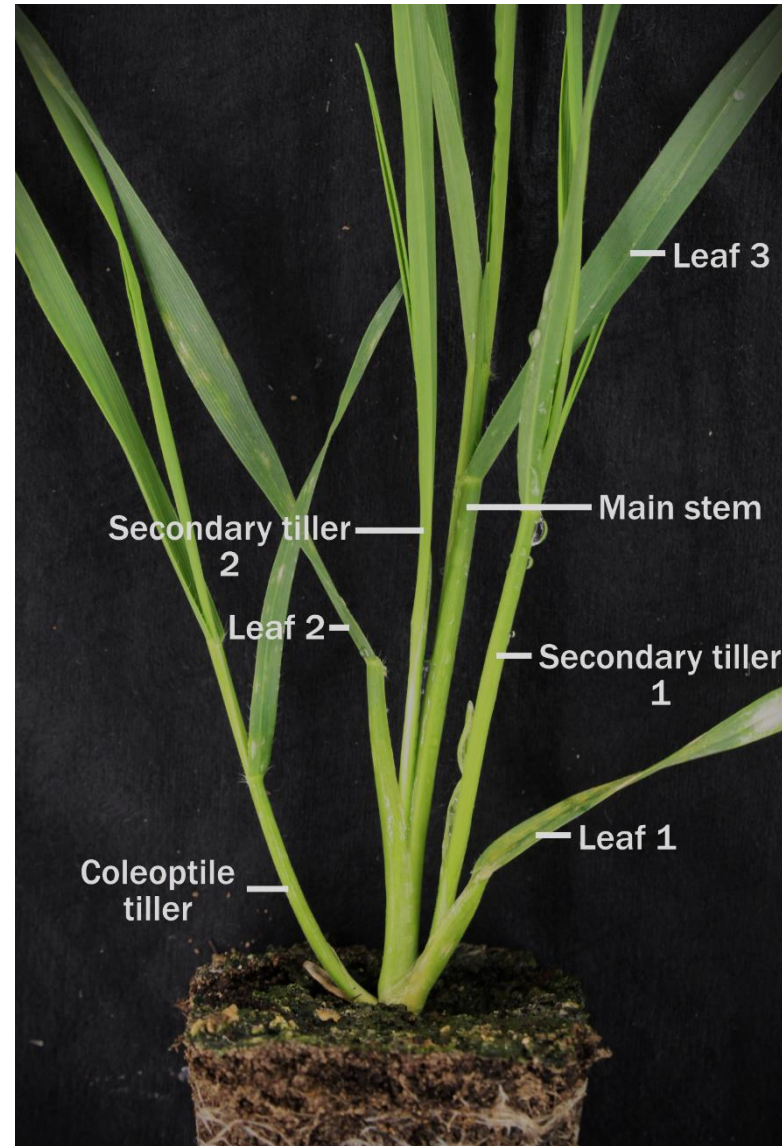
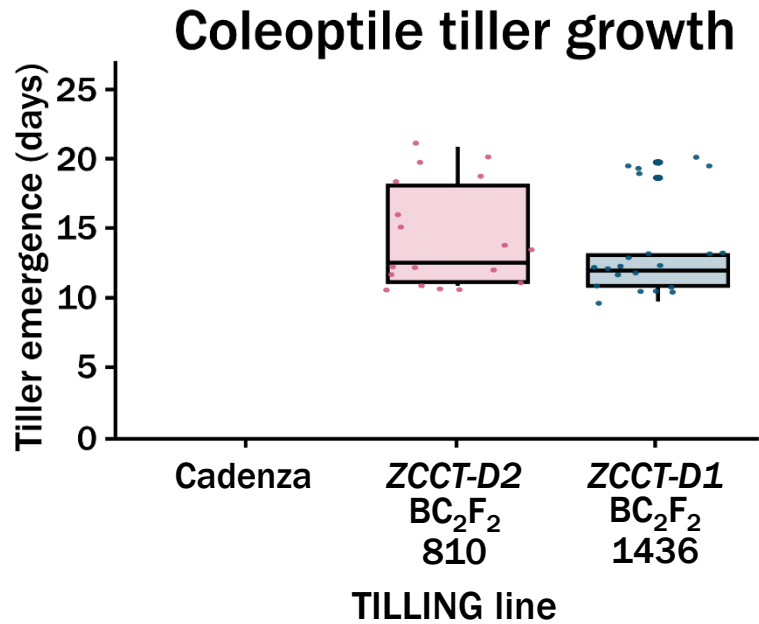
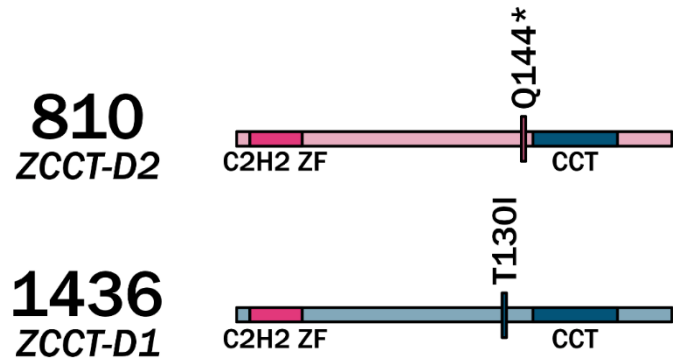
ZCCT-A2



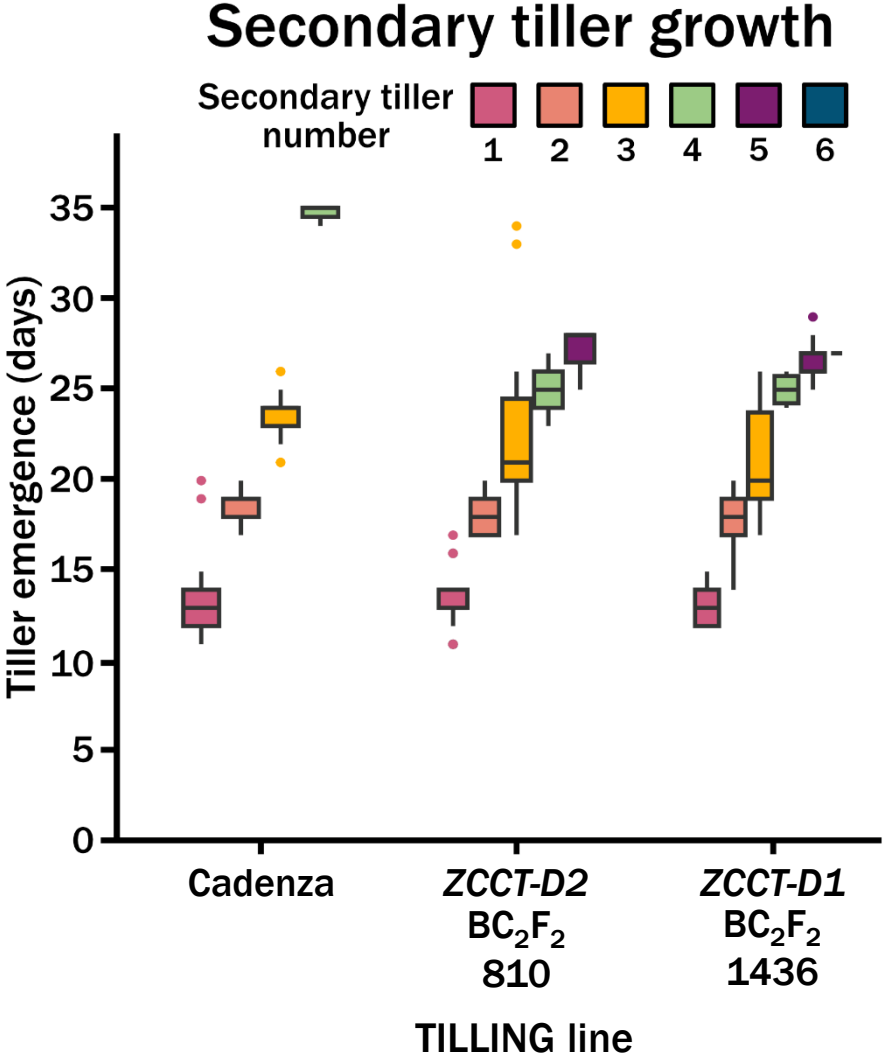
# VRN1 does not interact with all the ZCCTs



# ZCCT mutants have an increased number of coleoptile tillers



# ZCCT mutants have increased early tillering

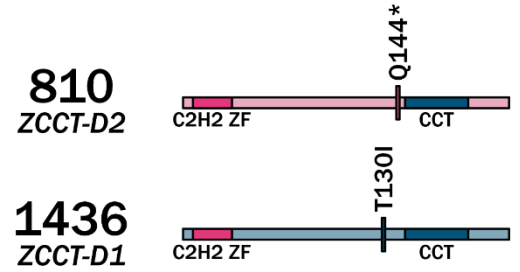


Cadenza

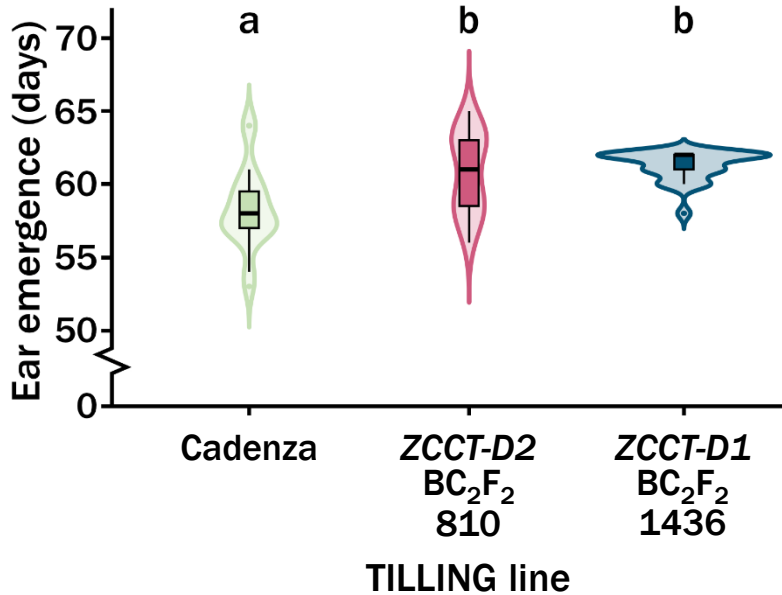


ZCCT-D2  
(BC<sub>2</sub>F<sub>2</sub> 810)

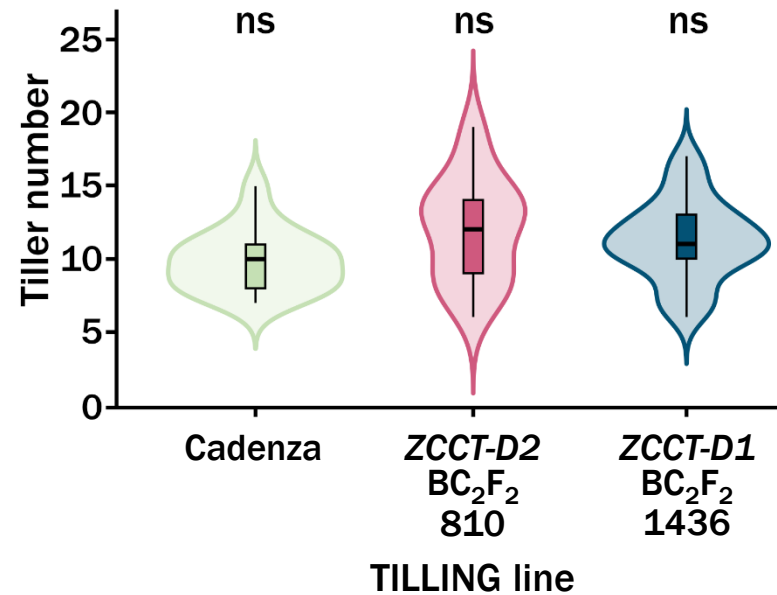
# ZCCT mutants have a slight delay in flowering time



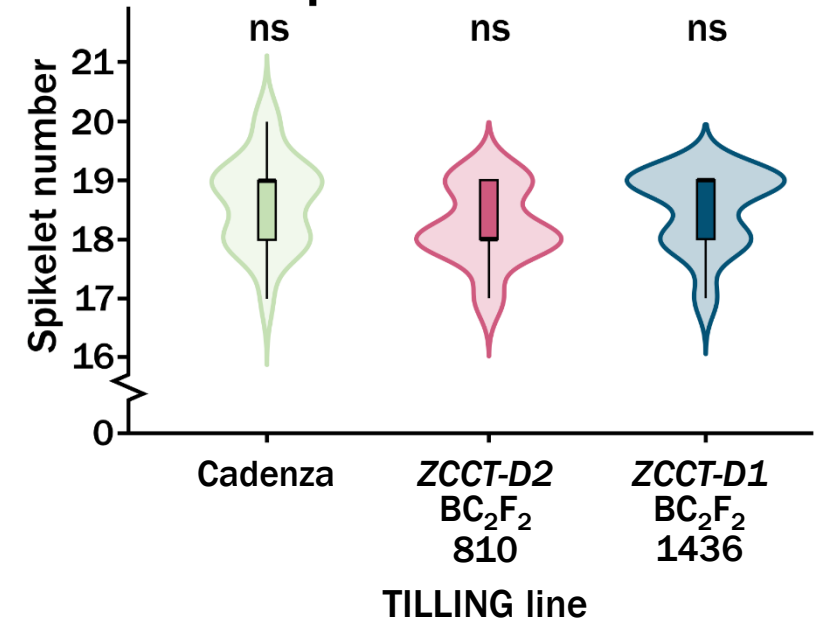
## Flowering time



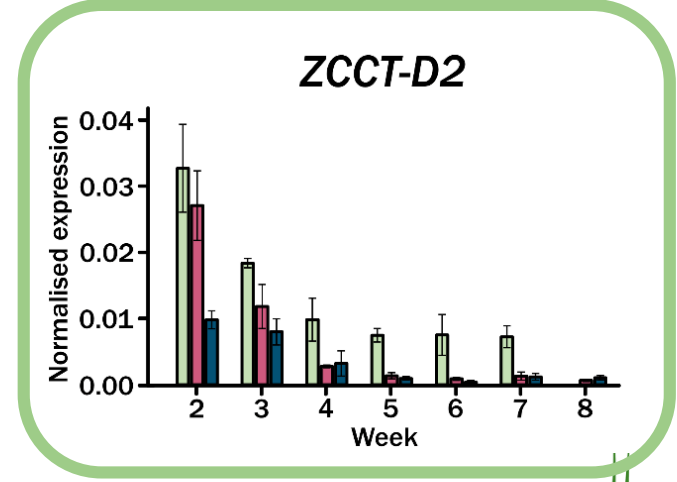
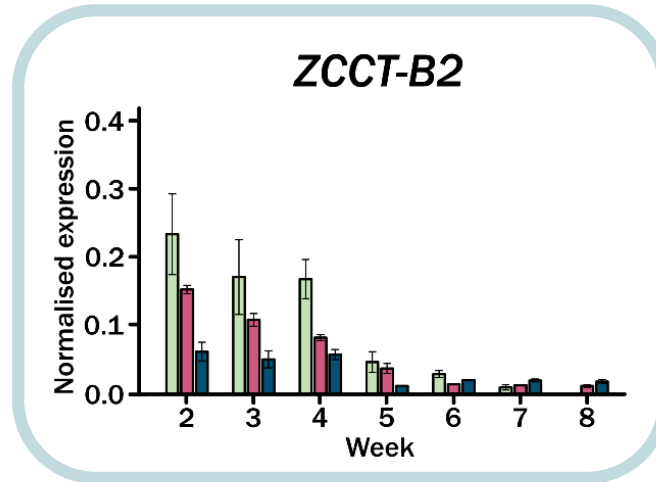
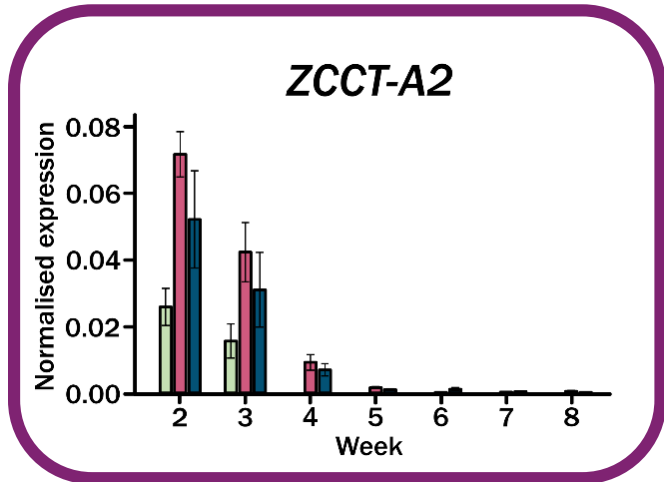
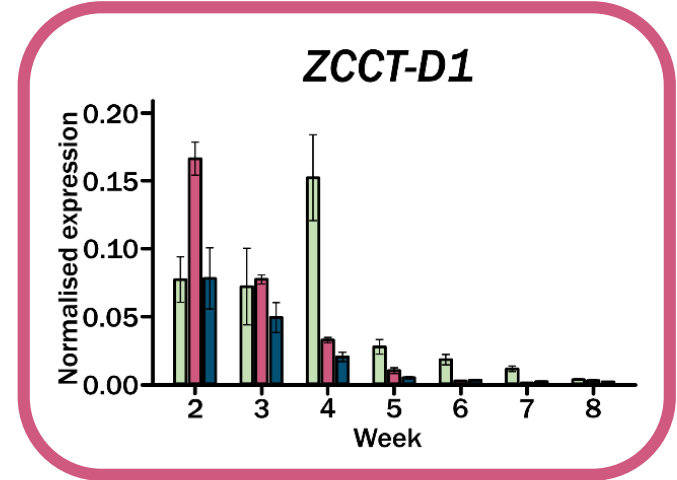
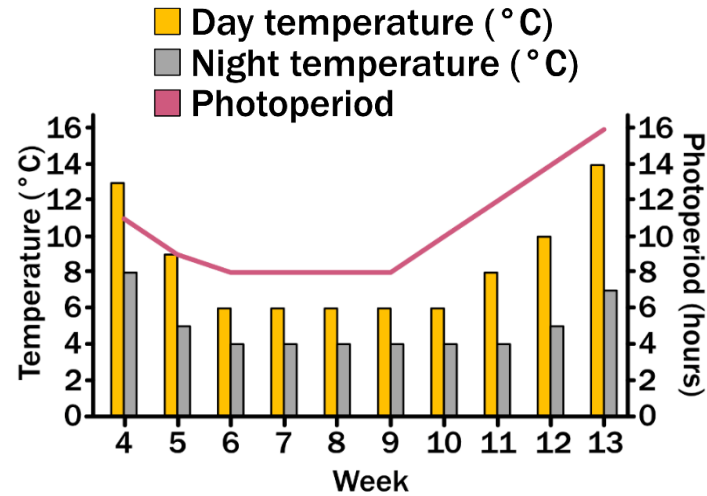
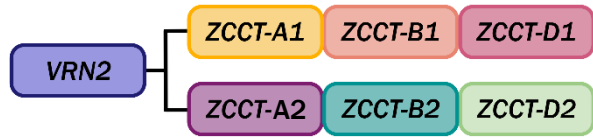
## Tiller number



## Spikelet number



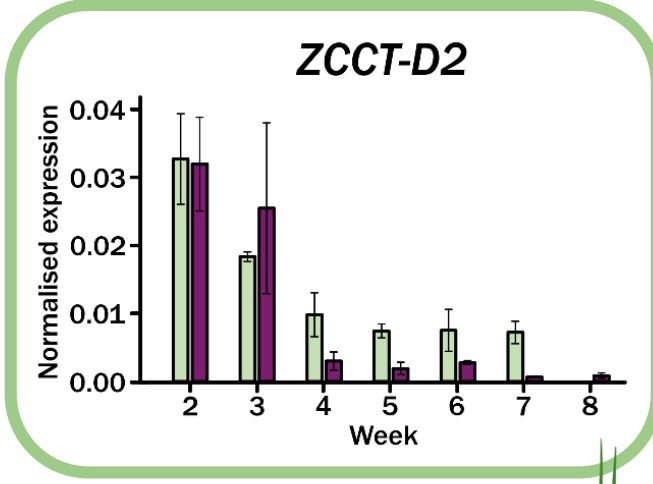
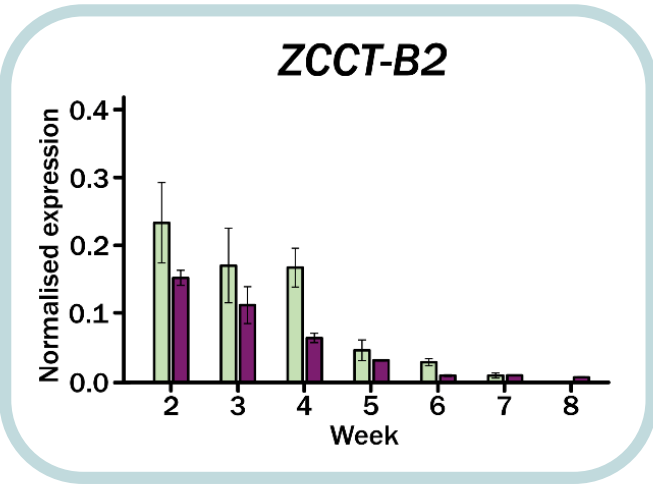
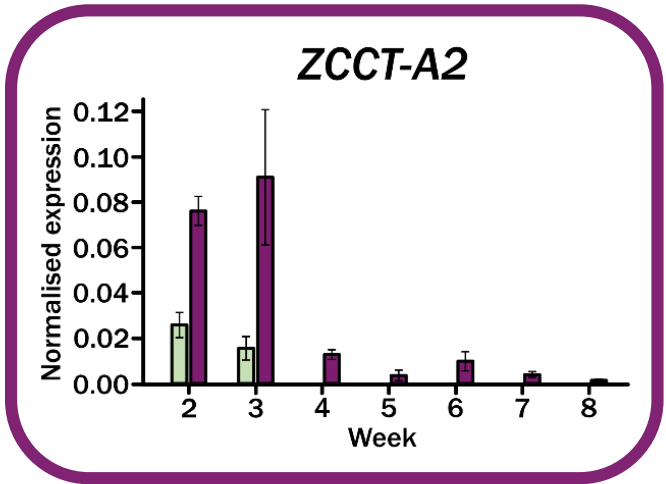
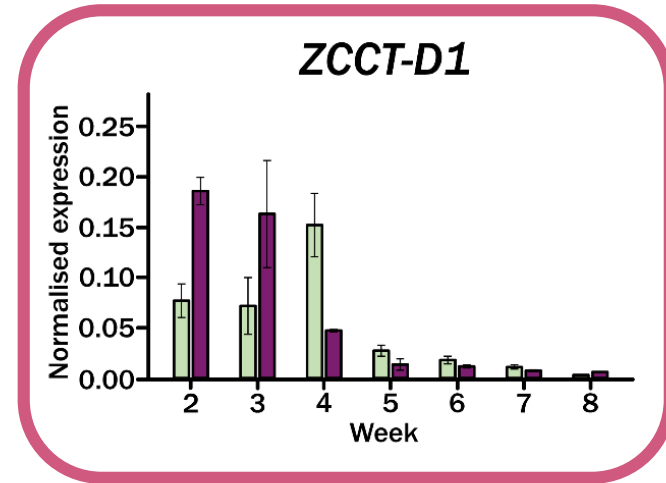
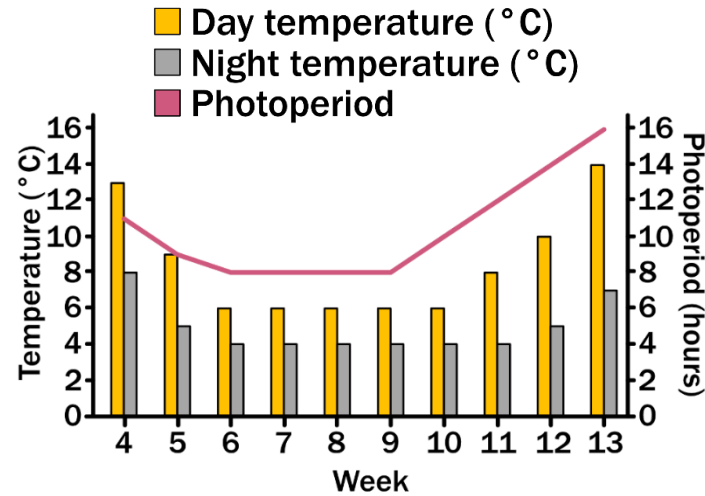
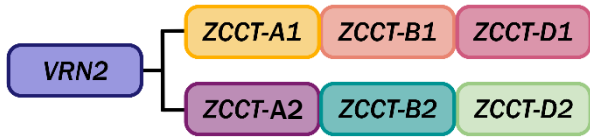
# Mutants in *ZCCT-D1* and *ZCCT-D2* show increased expression of *ZCCT-A2*



■ Cadenza  
■ *ZCCT-D2* (810)  
■ *ZCCT-D1* (1436)

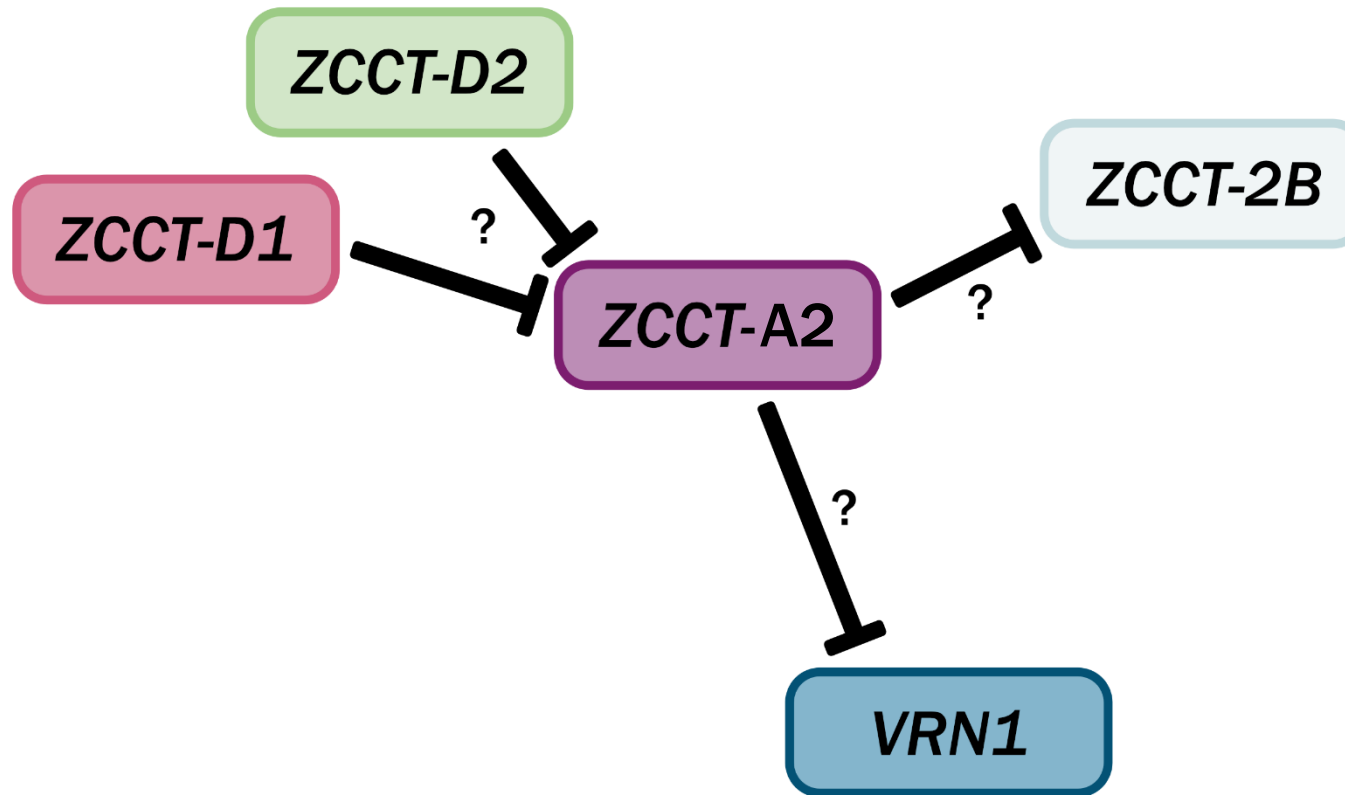


# Transgenic overexpression of *ZCCT-A2* also alters expression of *ZCCT-D1*



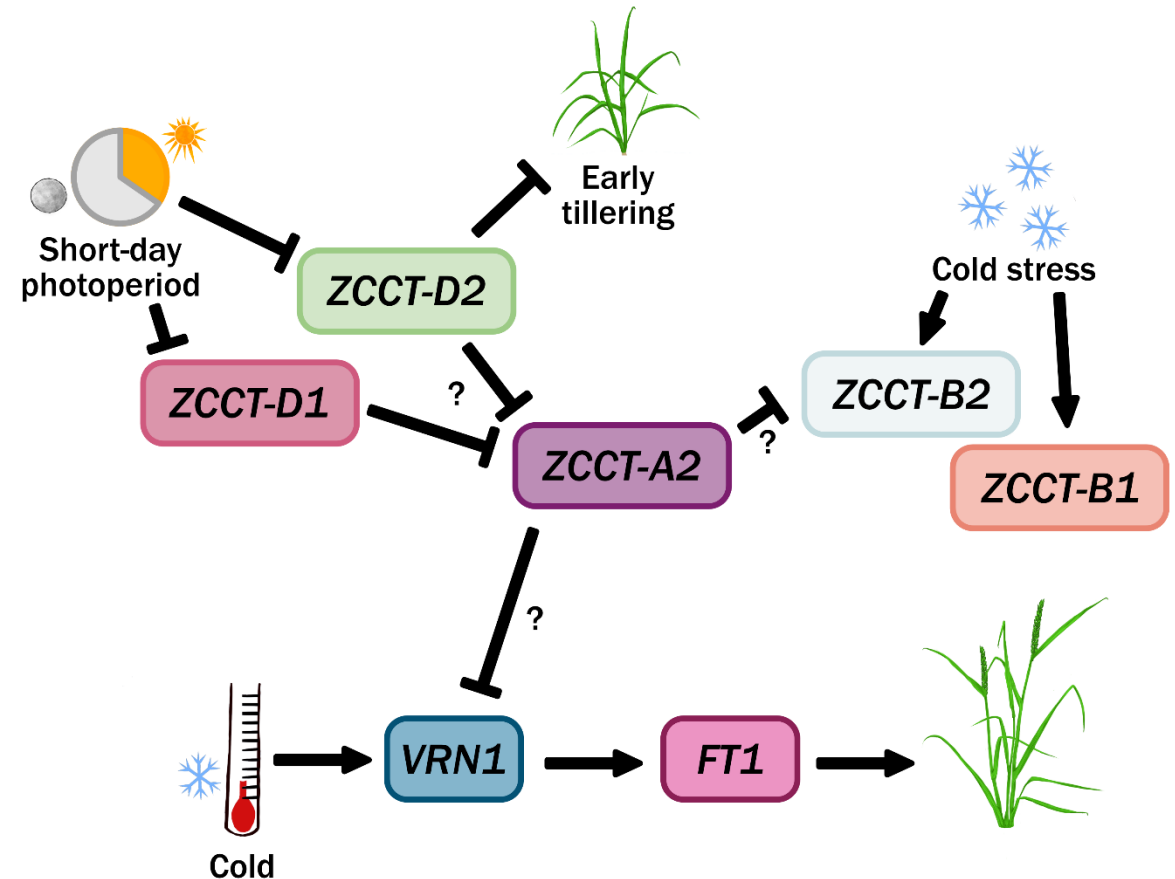
■ Cadenza  
■ ZCCT-A2-OE (CTA175.2)

# The *ZCCT* genes may regulate each other as a network



# Conclusions

- *VRN2* expression patterns are not always reflective of a vernalisation-responsive floral repressor
- There is potentially an additional developmental role for *VRN2*
- Not all *ZCCT* genes are behaving the same and likely form a regulatory network



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