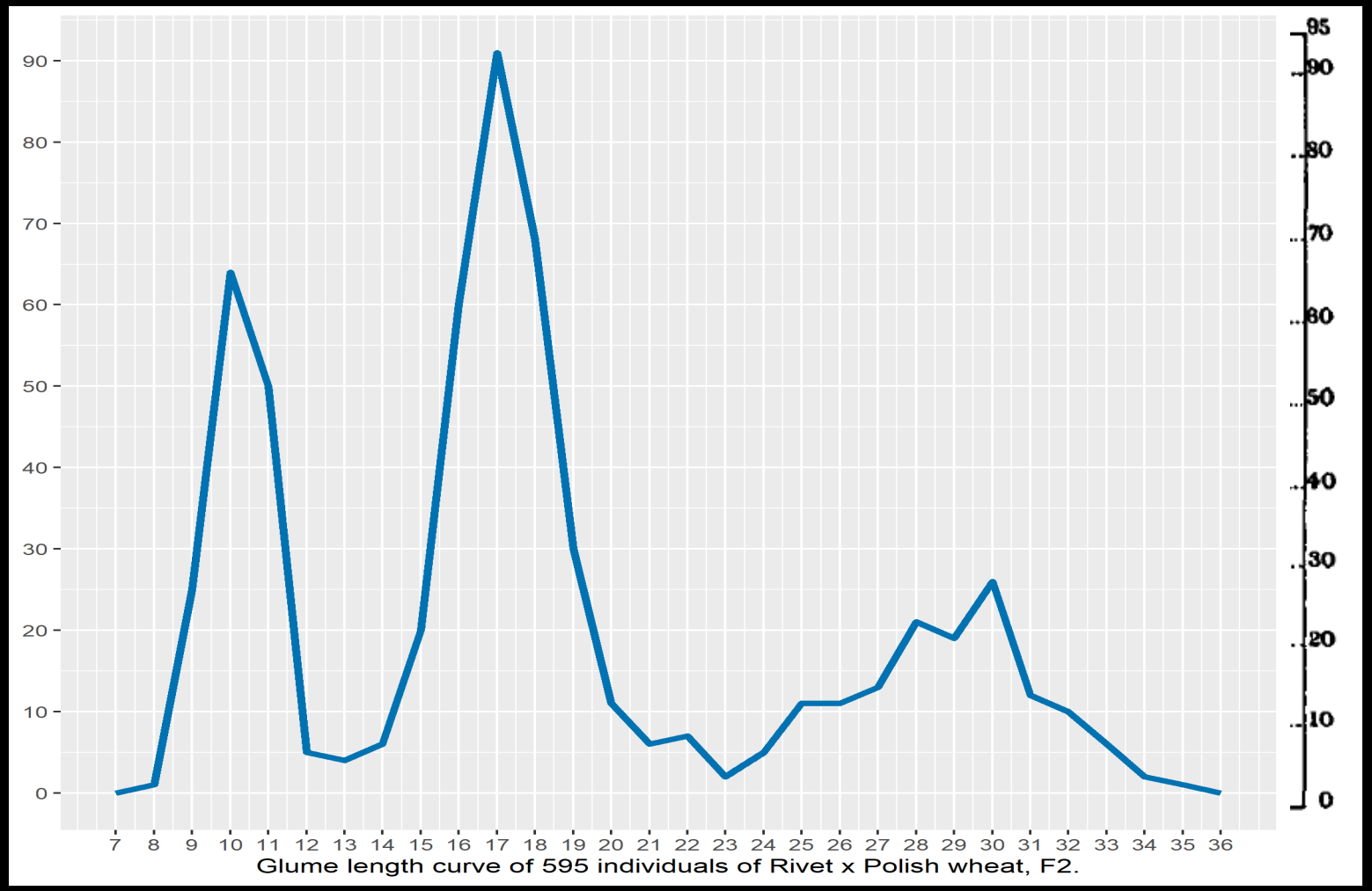
John Innes Centre

I W G S C

Solving a 100-year old mystery: Cloning the *P1* locus in *Triticum polonicum*

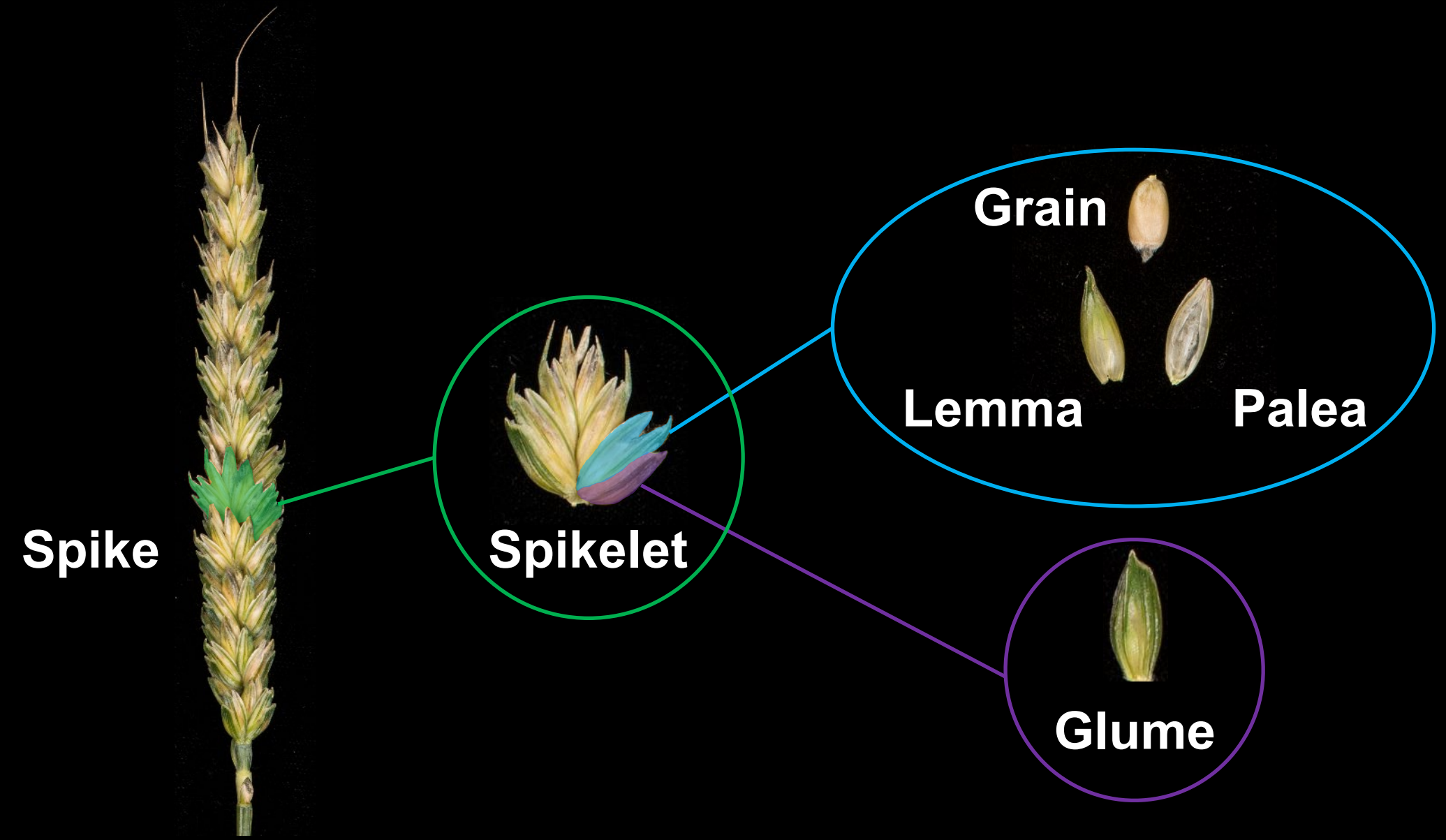
The *P1* locus of *Triticum polonicum* confers long glumes and grains

Vilmorin, 1880



Biffen 1905; Matsumura, 1950; Watanabe *et al.,* 1996; Kosuge *et al.,* 2010; Okamoto and Takumi, 2013

Short Terminology



Spikelet composition

Near Isogenic Lines (NILs)

T. polonicum

x

T. aestivum
cv. Paragon

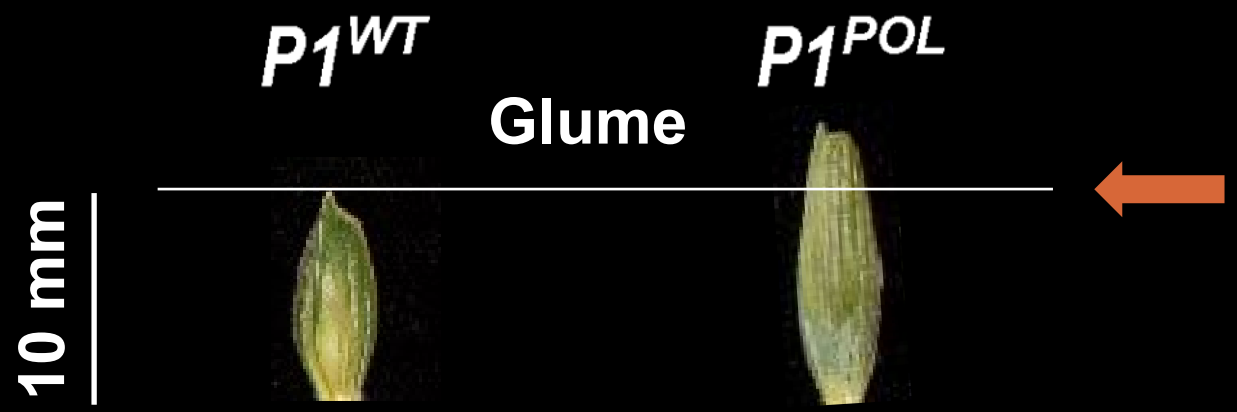
→ Backcross by phenotype (long glumes)

↓

BC 4 / BC 6 NILs

James Simmonds

Near Isogenic Lines (NILs)



Phenotypic effects of *P1* in field-grown NILs

$P1^{WT}$ $P1^{POL}$

+5.0%*** Grain length

+5.5%*** Thousand Grain Weight

+0.5%NS Yield

6 environments over 5 years; all data in Adamski *et al.,* 2021

Phenotypic effects of *P1* in field-grown NILs

$P1^{WT}$ $P1^{POL}$

+6 cm*** Crop Height

+1.6cm ** Spike length

-0.8 days*** Time to Heading

Breeder's toolkit

DESIGNING FUTURE WHEAT

6 environments over 5 years; all data in Adamski *et al.*, 2021

Mapping the *P1* locus - BC4 recombinants

Markers ordered via POPSEQ (genetic; cM)

POPSEQ [cM]	98.56	-	99.25	99.82	100.43	102.14	107.1
Axiom SNP	BS00022435	Phenotype	BS00022469	BS00022306	BS00021964	BS00033578	BS00077445
Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Polonicum	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol

Mascher *et al.*, 2013

Mapping the *P1* locus - BC4 recombinants

Markers ordered via POPSEQ (genetic; cM)

POPSEQ [cM]	98.56	-	99.25	99.82	100.43	102.14	107.1
Axiom SNP	BS00022435	Phenotype	BS00022469	BS00022306	BS00021964	BS00033578	BS00077445
Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_01	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_02	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_03	Paragon	Paragon	Paragon	T-pol	T-pol	Paragon	T-pol
Rec_04	Paragon	Paragon	T-pol	T-pol	T-pol	T-pol	T-pol
Rec_05	Paragon	Paragon	T-pol	T-pol	T-pol	T-pol	Paragon
Rec_06	Paragon	Paragon	T-pol	T-pol	T-pol	T-pol	Paragon
Rec_07	T-pol	T-pol	Paragon	Paragon	Paragon	Paragon	T-pol
Rec_08	T-pol	T-pol	Paragon	Paragon	Paragon	Paragon	T-pol
Rec_09	T-pol	T-pol	T-pol	Paragon	Paragon	T-pol	T-pol
Rec_10	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol
Rec_11	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol
Polonicum	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol

Mapping the *P1* locus - BC4 recombinants

Markers ordered with RefSeqv0.4 (physical; bp)

Mapping the *P1* locus - BC4 recombinants

Markers ordered with RefSeqv0.4 (physical; bp)

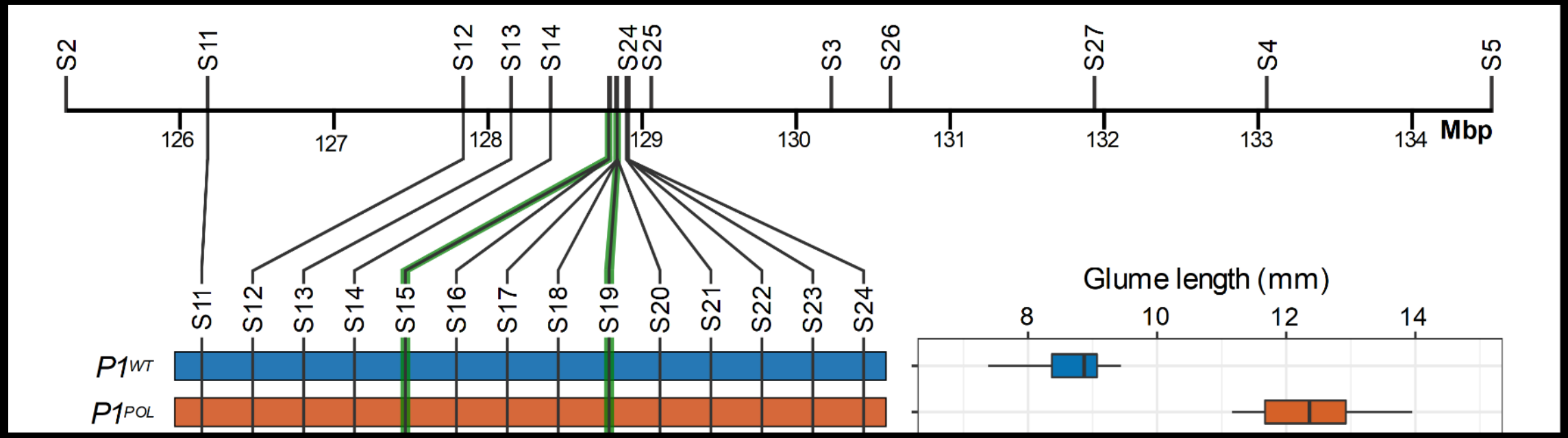
POPSEQ [cM]	99.82	100.43	99.25	102.14	98.56	-	107.1
Axiom SNP	BS00022306	BS00021964	BS00022469	BS00033578	BS00022435	-	BS00077445
RefSeqv0.4 [bp]	117,276,239	118,073,697	120,105,578	120,899,237	127,502,160	Phenotype	544,983,113

Mapping the *P1* locus - BC4 recombinants

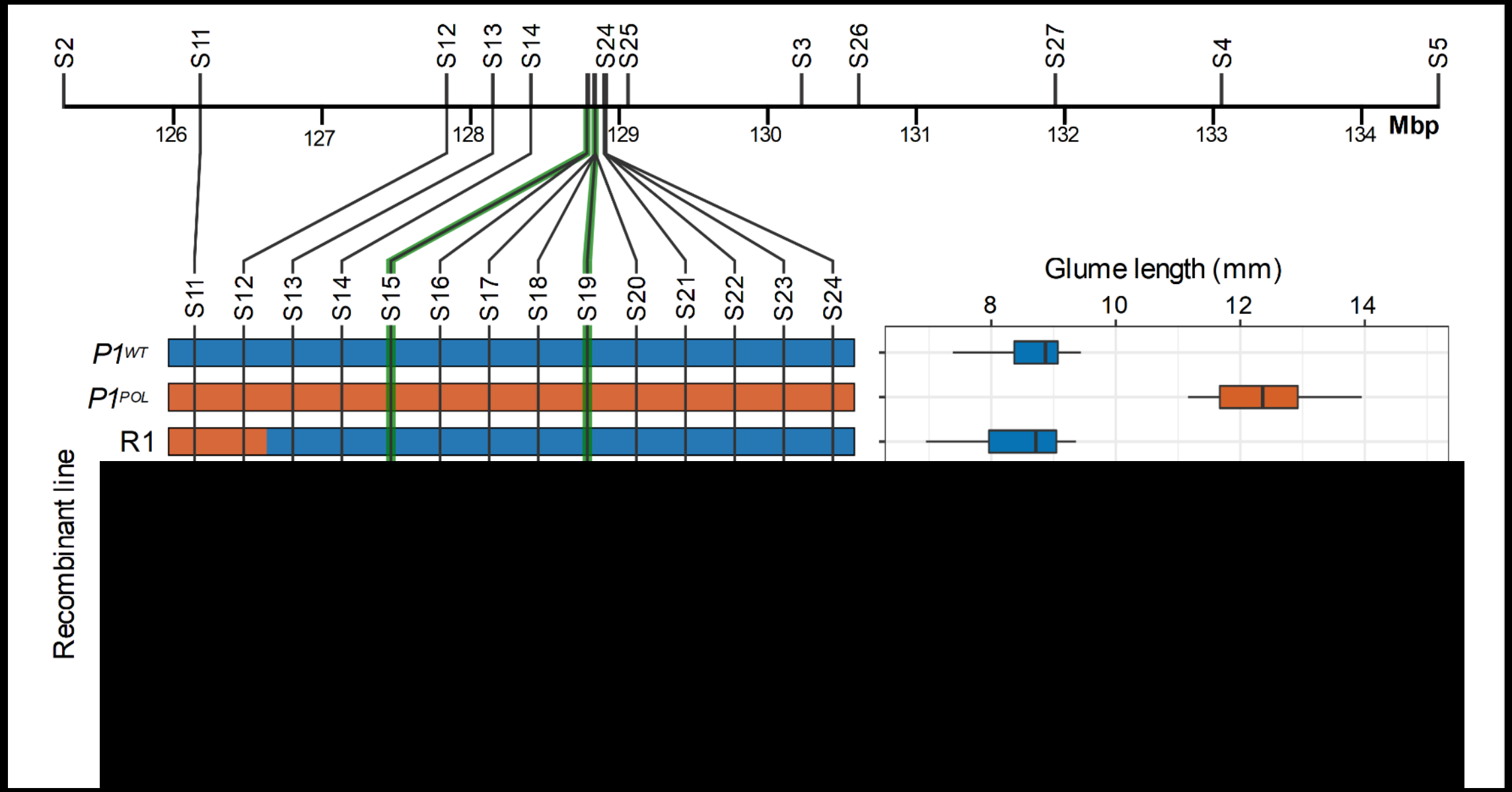
Markers ordered with RefSeqv0.4 (physical; bp)

POPSEQ [cM]	99.82	100.43	99.25	102.14	98.56	-	107.1
Axiom SNP	BS00022306	BS00021964	BS00022469	BS00033578	BS00022435	-	BS00077445
RefSeqv0.4 [bp]	117,276,239	118,073,697	120,105,578	120,899,237	127,502,160	Phenotype	544,983,113
Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_01	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_02	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon	Paragon
Rec_03	T-pol	T-pol	Paragon	Paragon	Paragon	Paragon	T-pol
Rec_04	T-pol	T-pol	T-pol	T-pol	Paragon	Paragon	T-pol
Rec_05	T-pol	T-pol	T-pol	T-pol	Paragon	Paragon	Paragon
Rec_06	T-pol	T-pol	T-pol	T-pol	Paragon	Paragon	Paragon
Rec_07	Paragon	Paragon	Paragon	Paragon	T-pol	T-pol	T-pol
Rec_08	Paragon	Paragon	Paragon	Paragon	T-pol	T-pol	T-pol
Rec_09	Paragon	Paragon	T-pol	T-pol	T-pol	T-pol	T-pol
Rec_10	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol
Rec_11	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol
Polonicum	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol	T-pol

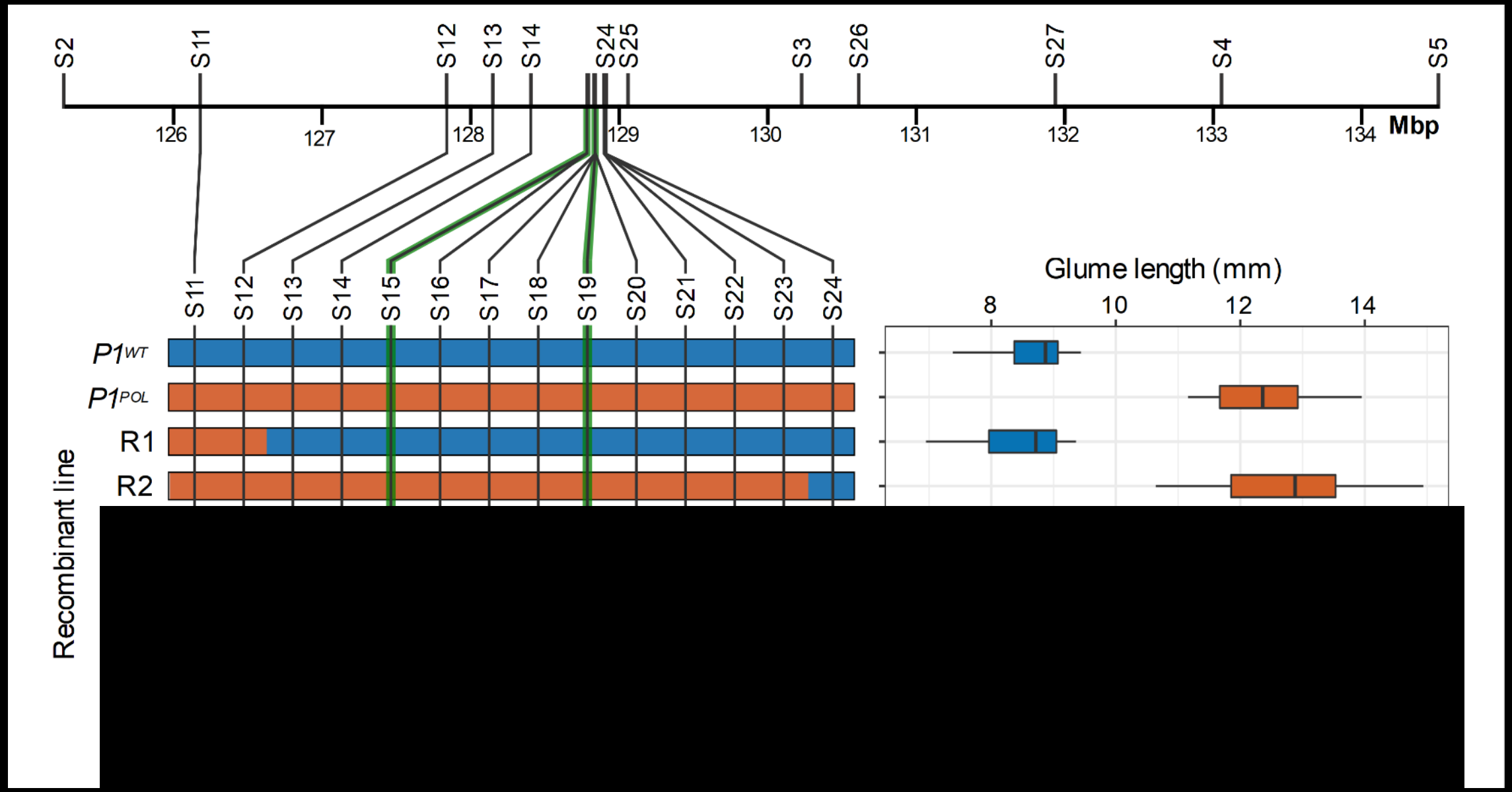
Mapping the *P1* locus - BC6 recombinants



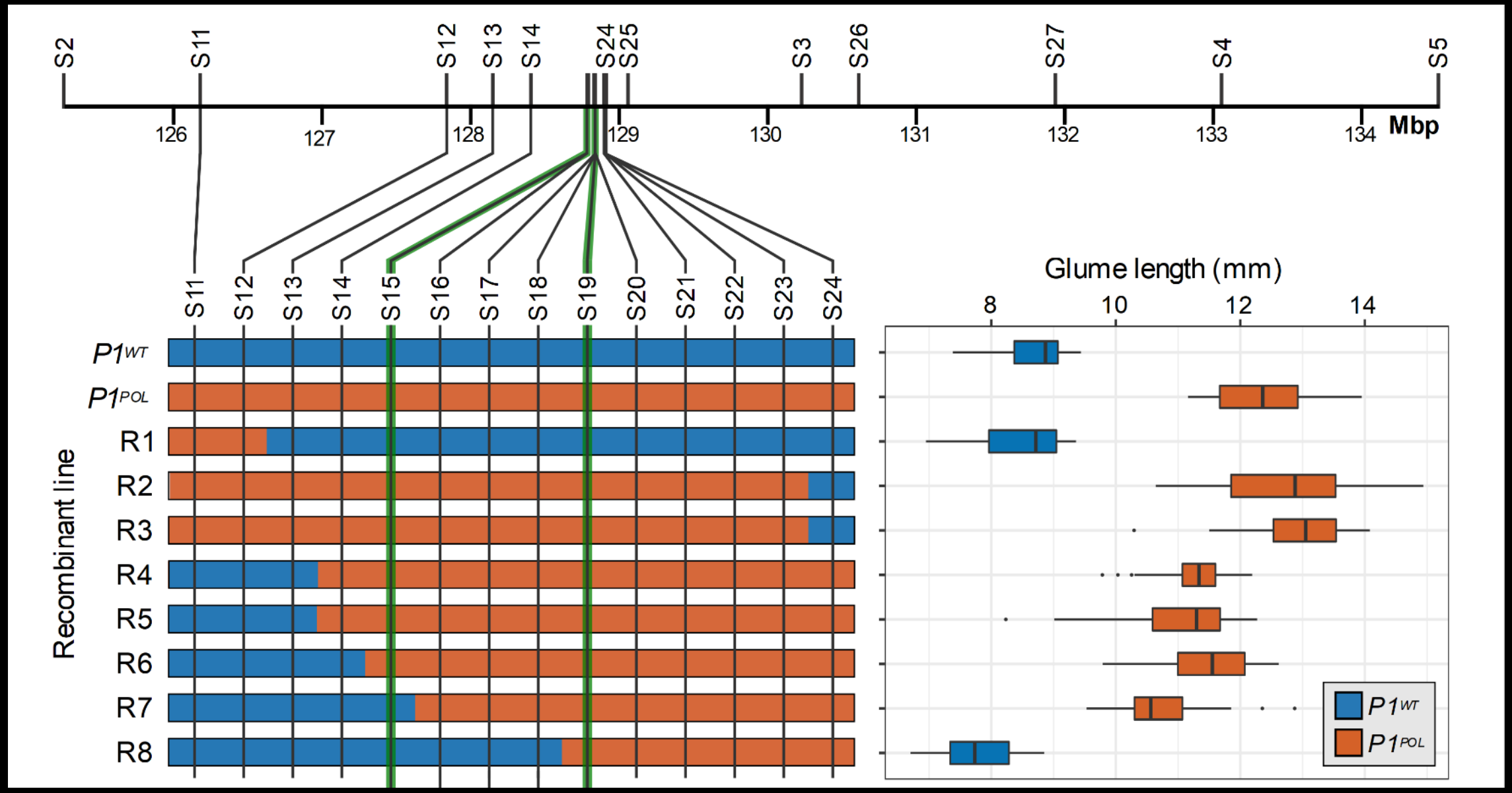
Mapping the *P1* locus - BC6 recombinants



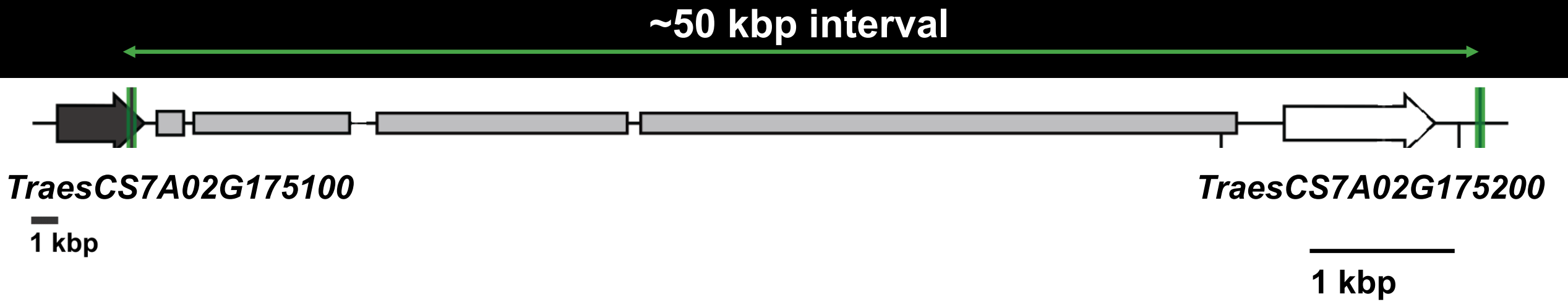
Mapping the *P1* locus - BC6 recombinants



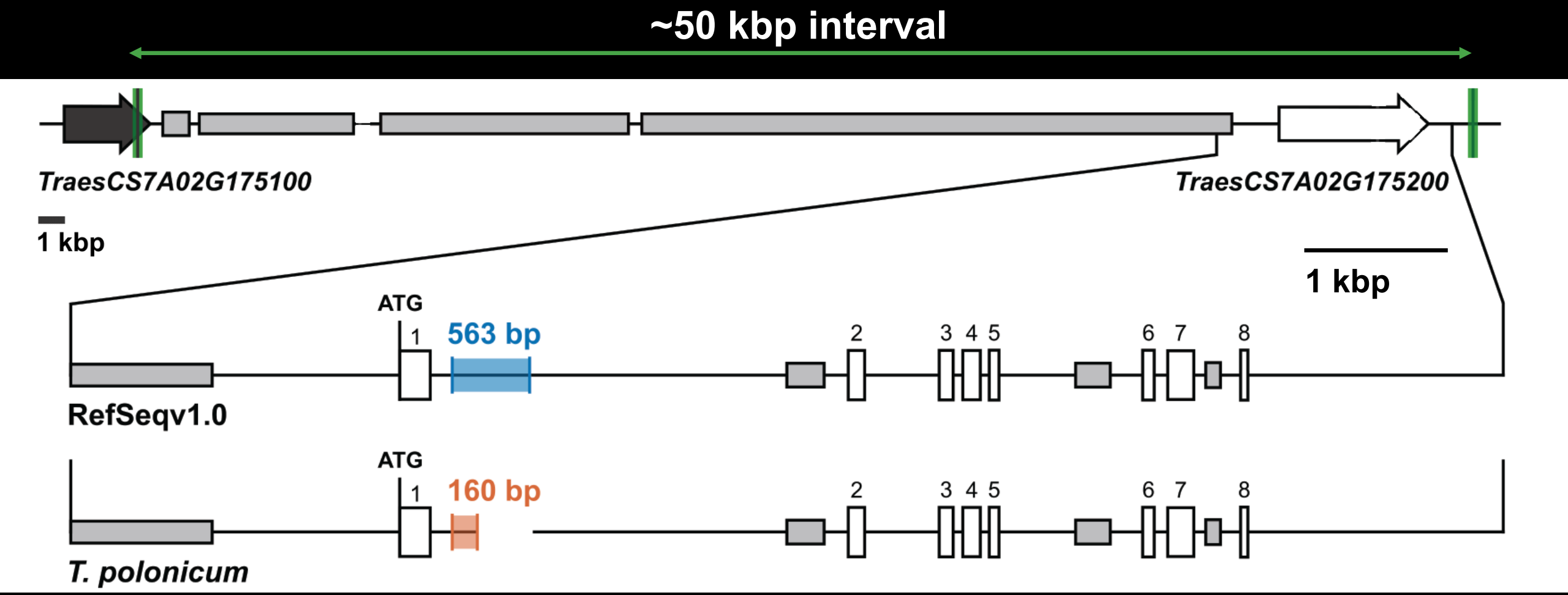
Mapping the *P1* locus - BC6 recombinants



Fine-mapping the *P1* locus



Fine-mapping the *P1* locus



Adamski *et al.*, 2021; Liu *et al.*, 2021; Chai *et al.*, 2021; Xiao *et al.*, 2021

Allele classification

TraesCS7A02G175200 **is a** ***Short Vegetative Phase*** **(*SVP*) homolog**

Allele classification

TraesCS7A02G175200 **is a** ***Short Vegetative Phase*** **(*SVP*) homolog**

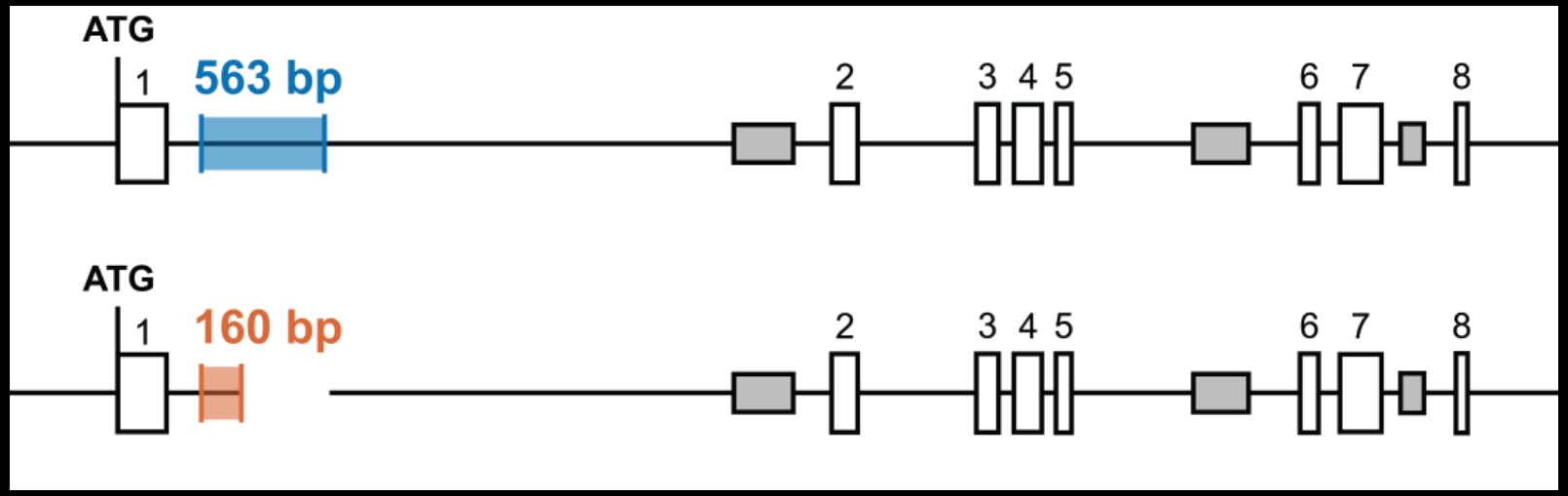
VEGETATIVE TO REPRODUCTIVE TRANSITION 2 **(*VRT2*)**

Kane *et al.*, 2005

Allele classification

***TraesCS7A02G175200* is a *Short Vegetative Phase* (*SVP*) homolog**

***VEGETATIVE TO REPRODUCTIVE TRANSITION 2* (*VRT2*)**



***VRT-A2a* allele (wildtype)**

***VRT-A2b* allele (*polonicum*)**

Adamski *et al.*, 2021; Liu *et al.*, 2021

Potential origin of the *VRT-A2b* allele

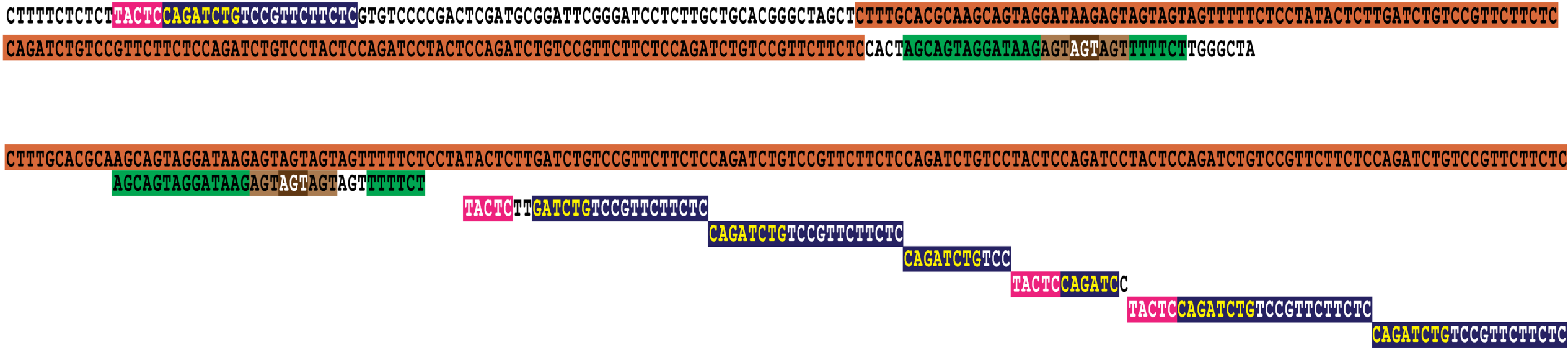
CTTTTCTCTCTTACTCCAGATCTGTCCGTTCTTCTCGTGTCCCCGACTCGATGCGGATTCGGGATCCTCTTGCTGCACGGGCTAGCTCTTTGCACGCAAGCAGTAGGATAAGAGTAGTAGTAGTTTTTCTCCTATACTCTTGATCTGTCCGTTCTTCTC
CAGATCTGTCCGTTCTTCTCCAGATCTGTCCTACTCCAGATCCTACTCCAGATCTGTCCGTTCTTCTCCAGATCTGTCCGTTCTTCTCCACTAGCAGTAGGATAAGAGTAGTAGTTTTTCTTGGGCTA

Potential origin of the *VRT-A2b* allele

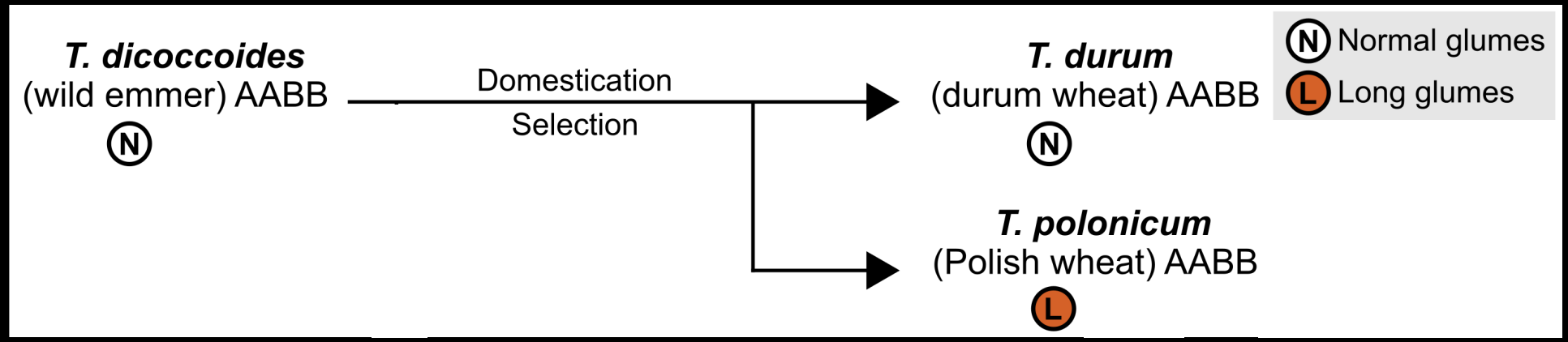
CTTTTCTCTCTTACTCCAGATCTGTCCGTTCTTCTCGTGTCCCCGACTCGATGCGGATTCGGGATCCTCTTGCTGCACGGGCTAGCTCTTTGCACGCAAGCAGTAGGATAAGAGTAGTAGTAGTTTTTCTCCTATACTCTTGATCTGTCCGTTCTTCTC

CAGATCTGTCCGTTCTTCTCCAGATCTGTCCTACTCCAGATCCTACTCCAGATCTGTCCGTTCTTCTCCAGATCTGTCCGTTCTTCTCCACTAGCAGTAGGATAAGAGTAGTAGTTTTTCTTGGGCTA

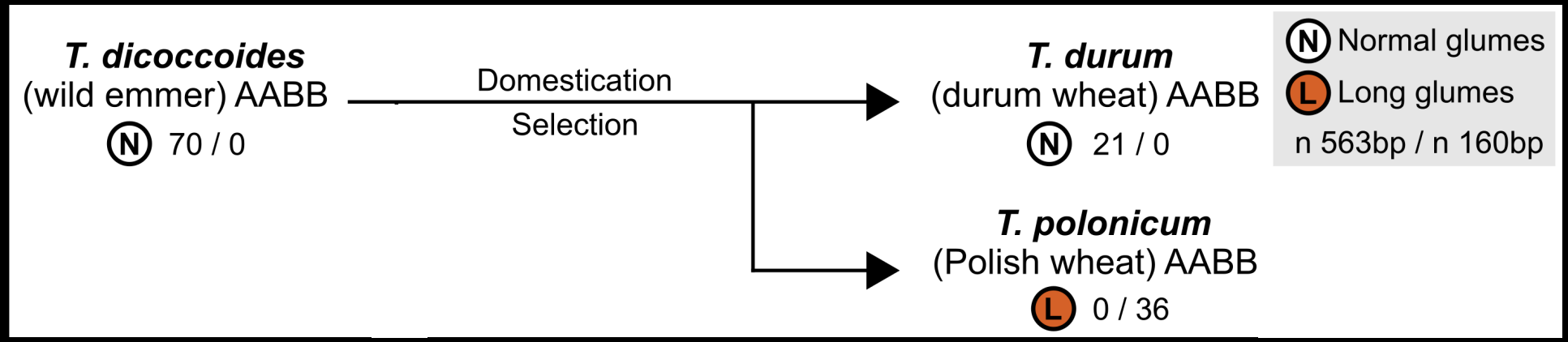
Potential origin of the *VRT-A2b* allele



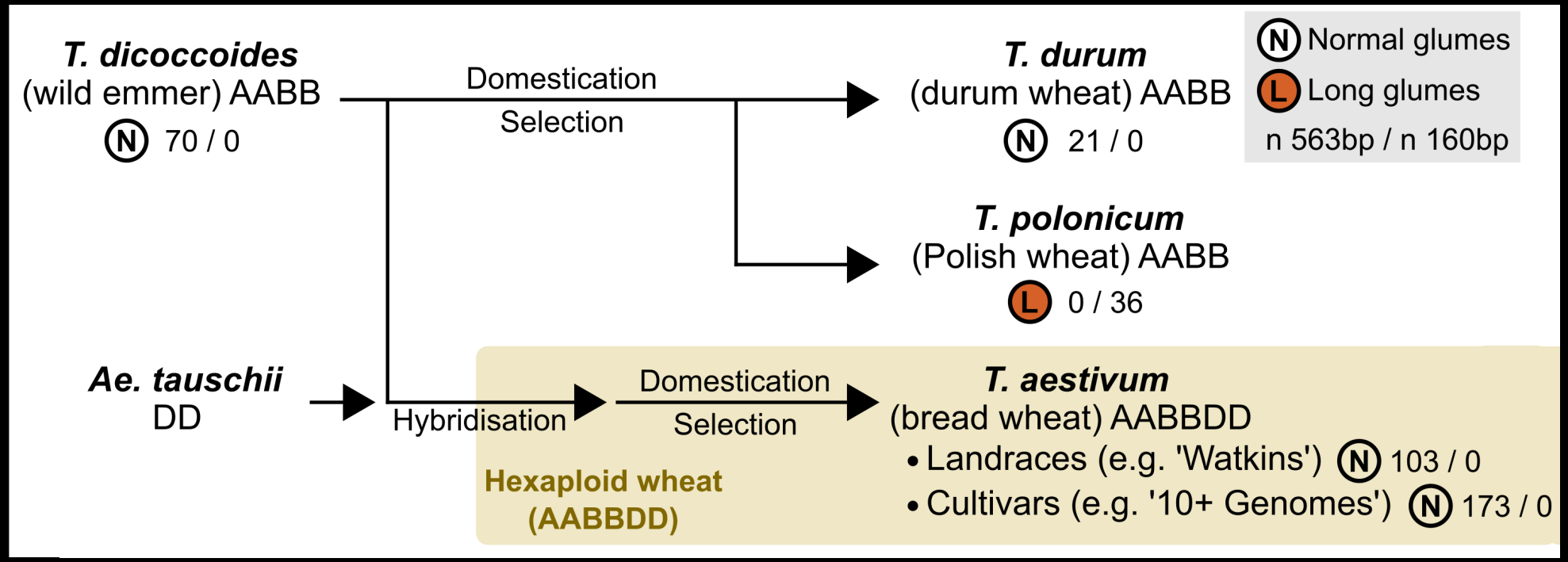
Distribution of *P1* allele



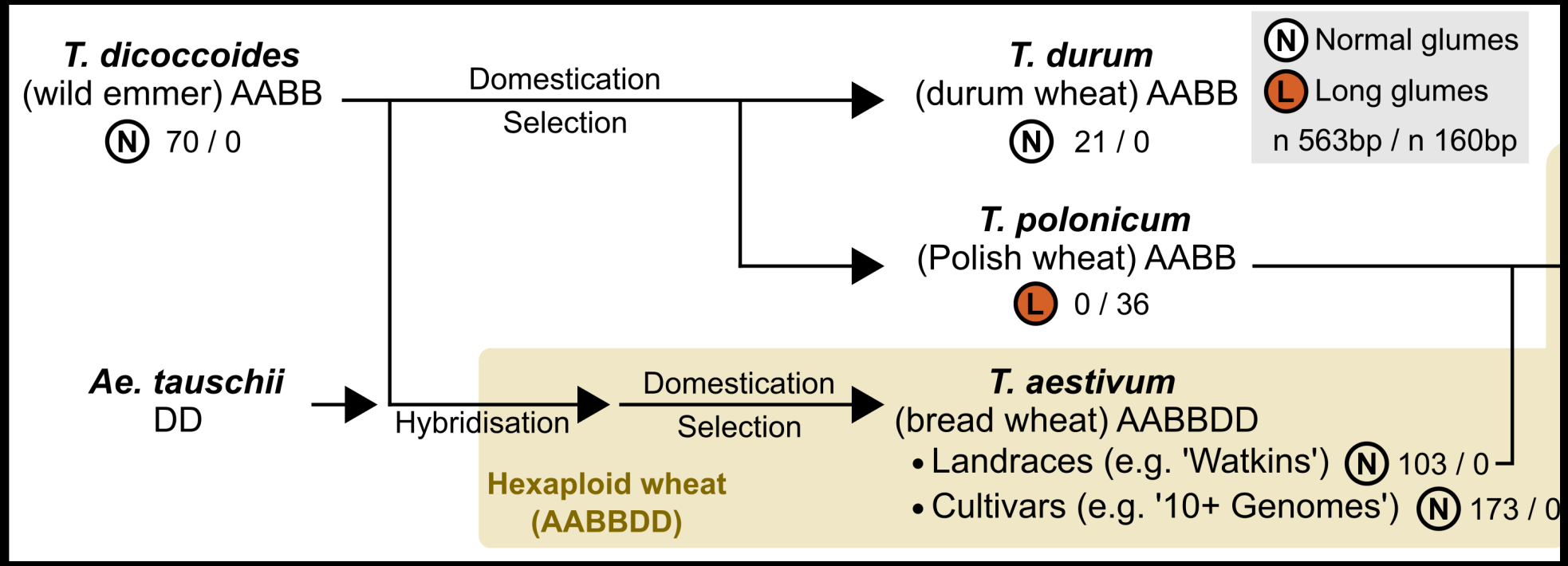
Distribution of *P1* allele

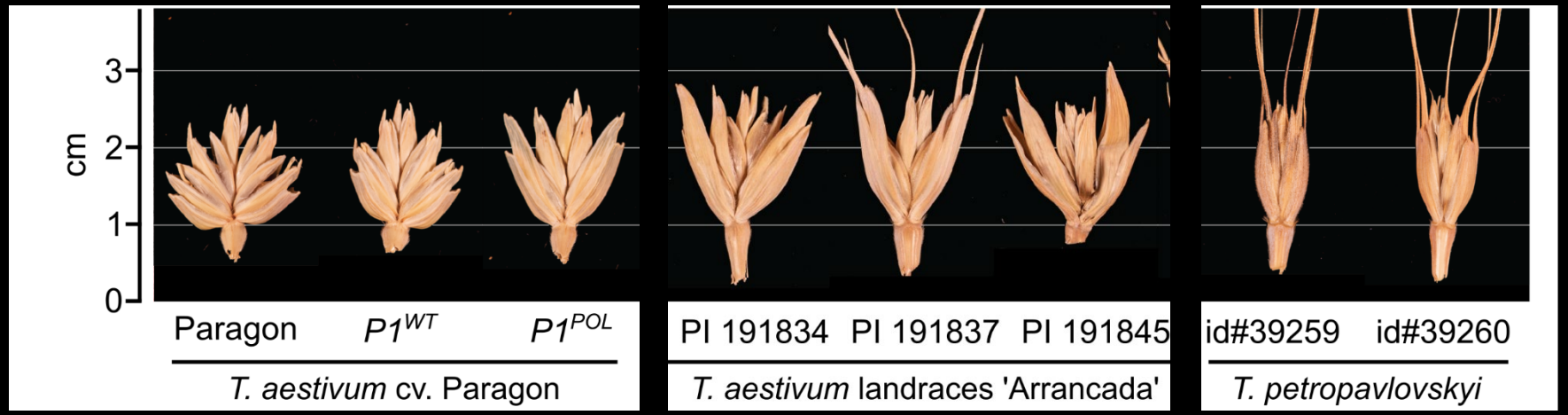


Distribution of *P1* allele



Distribution of *P1* allele





Distribution of *P1* allele

T. dicoccoides
(wild emmer) AABB
N 70 / 0

Domestication
Selection

T. durum
(durum wheat) AABB
N 21 / 0

N Normal glumes
L Long glumes
n 563bp / n 160bp

T. polonicum
(Polish wheat) AABB
L 0 / 36

***T. aestivum* 'Arrancada'**
AABBDD
L 0 / 7

T. petropavlovskyi
AABBDD
L 0 / 4

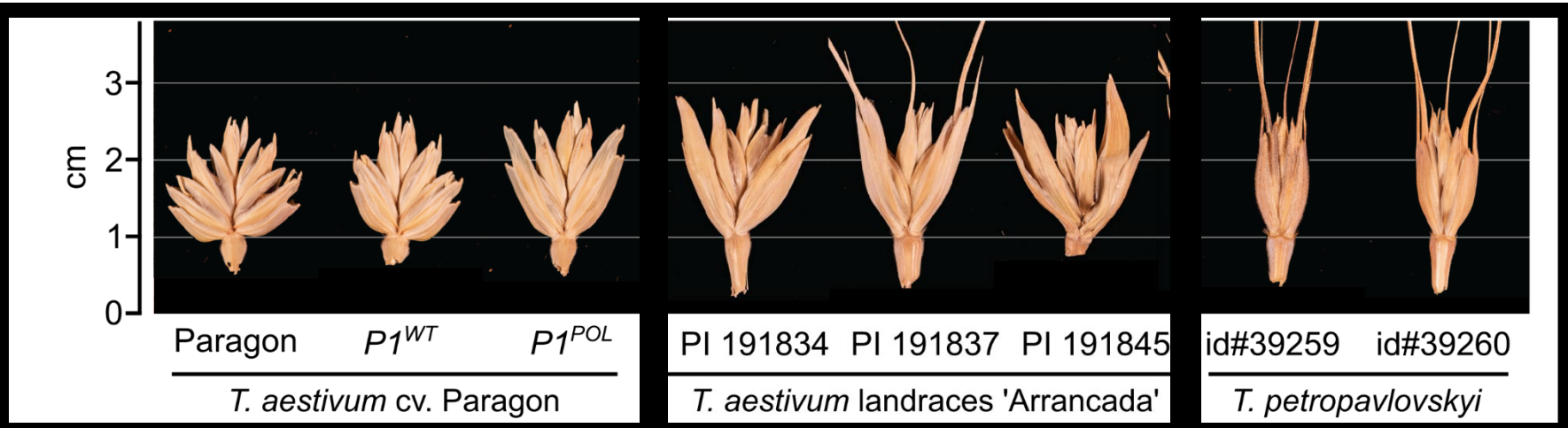
Ae. tauschii
DD

Hybridisation

Domestication
Selection

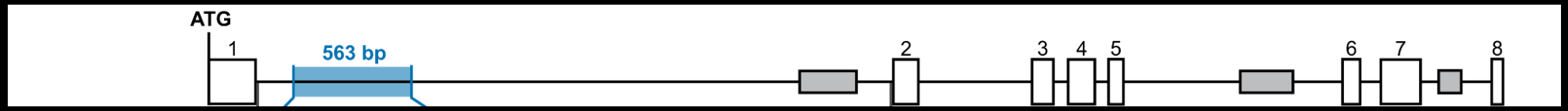
T. aestivum
(bread wheat) AABBDD
- Landraces (e.g. 'Watkins') N 103 / 0
- Cultivars (e.g. '10+ Genomes') N 173 / 0

Hexaploid wheat (AABBDD)

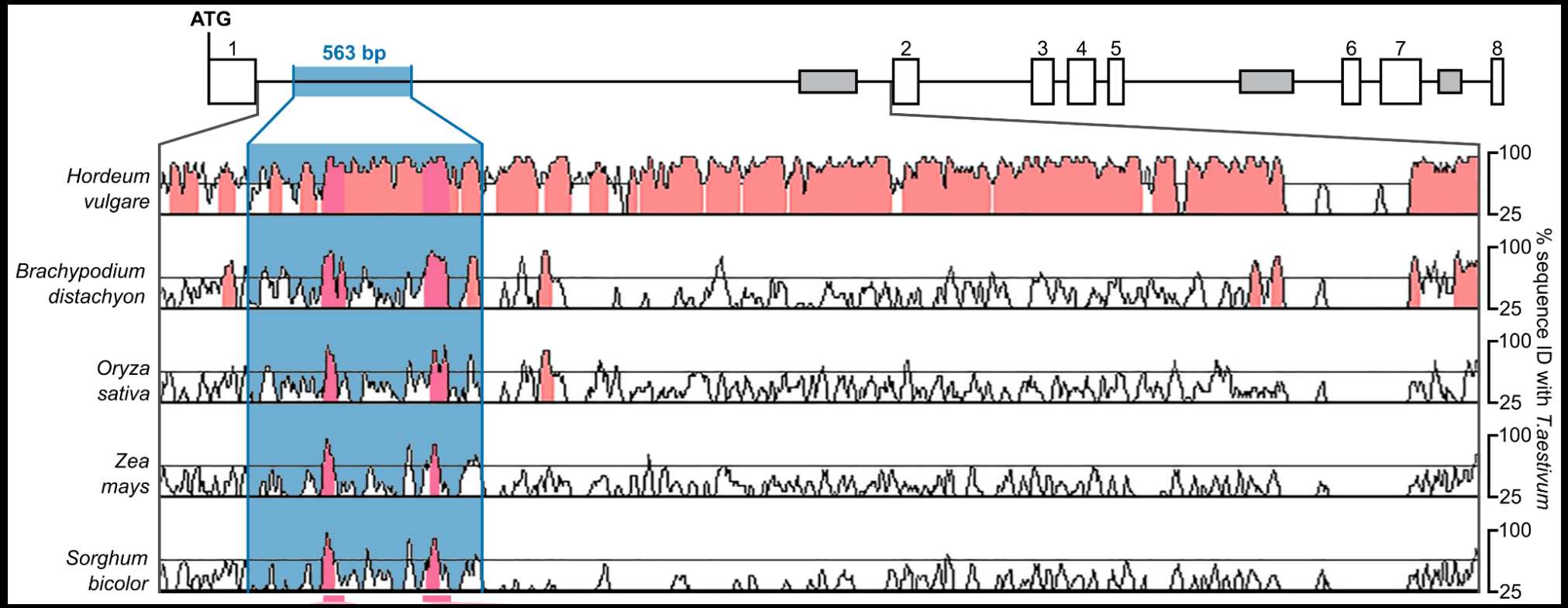


Adamski *et al.*, 2021
Liu *et al.*, 2021
Chai *et al.*, 2021
Xiao *et al.*, 2021

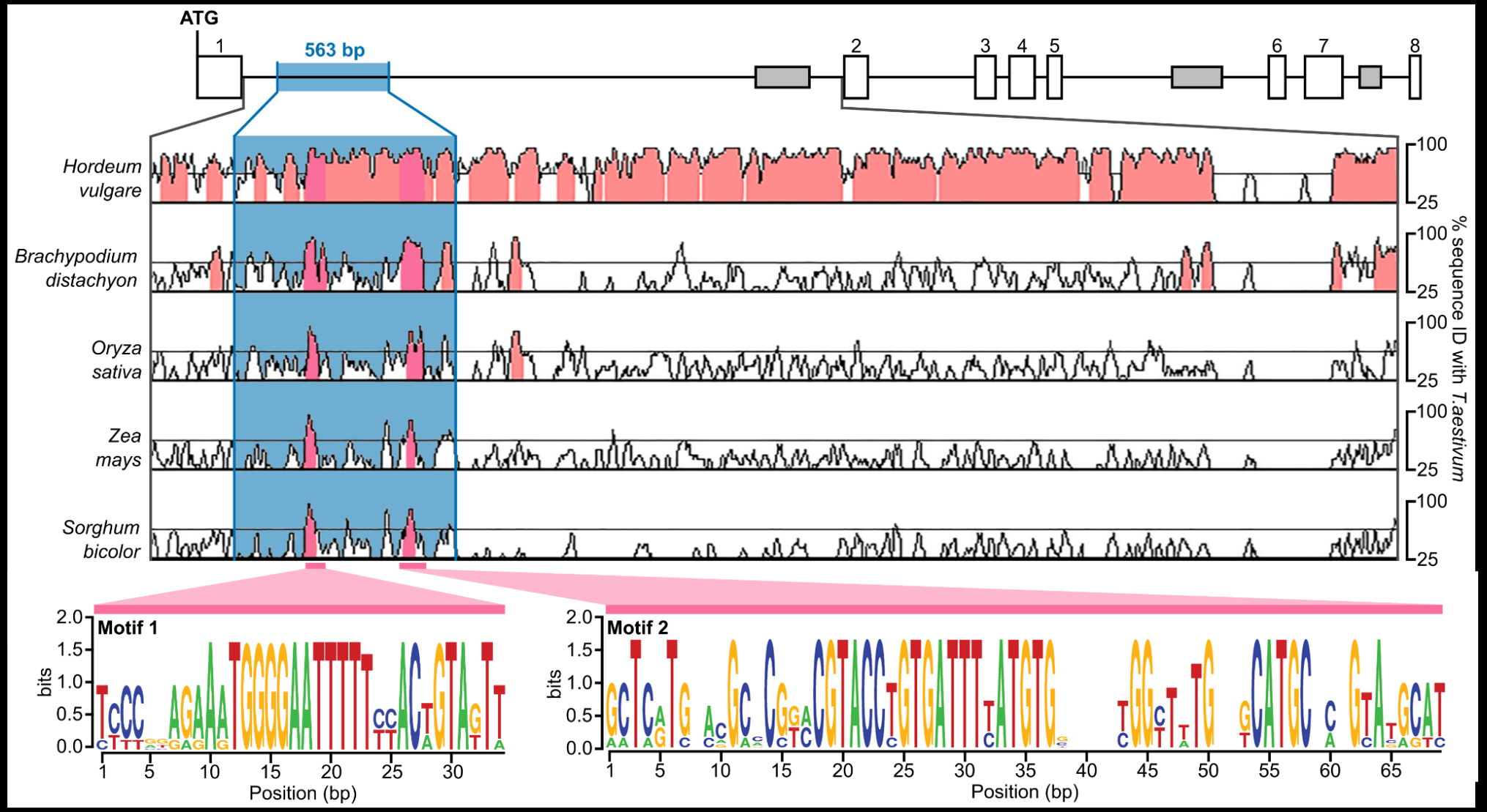
Conservation of *VRT-A2a* intron 1 sequence across grasses



Conservation of *VRT-A2a* intron 1 sequence across grasses

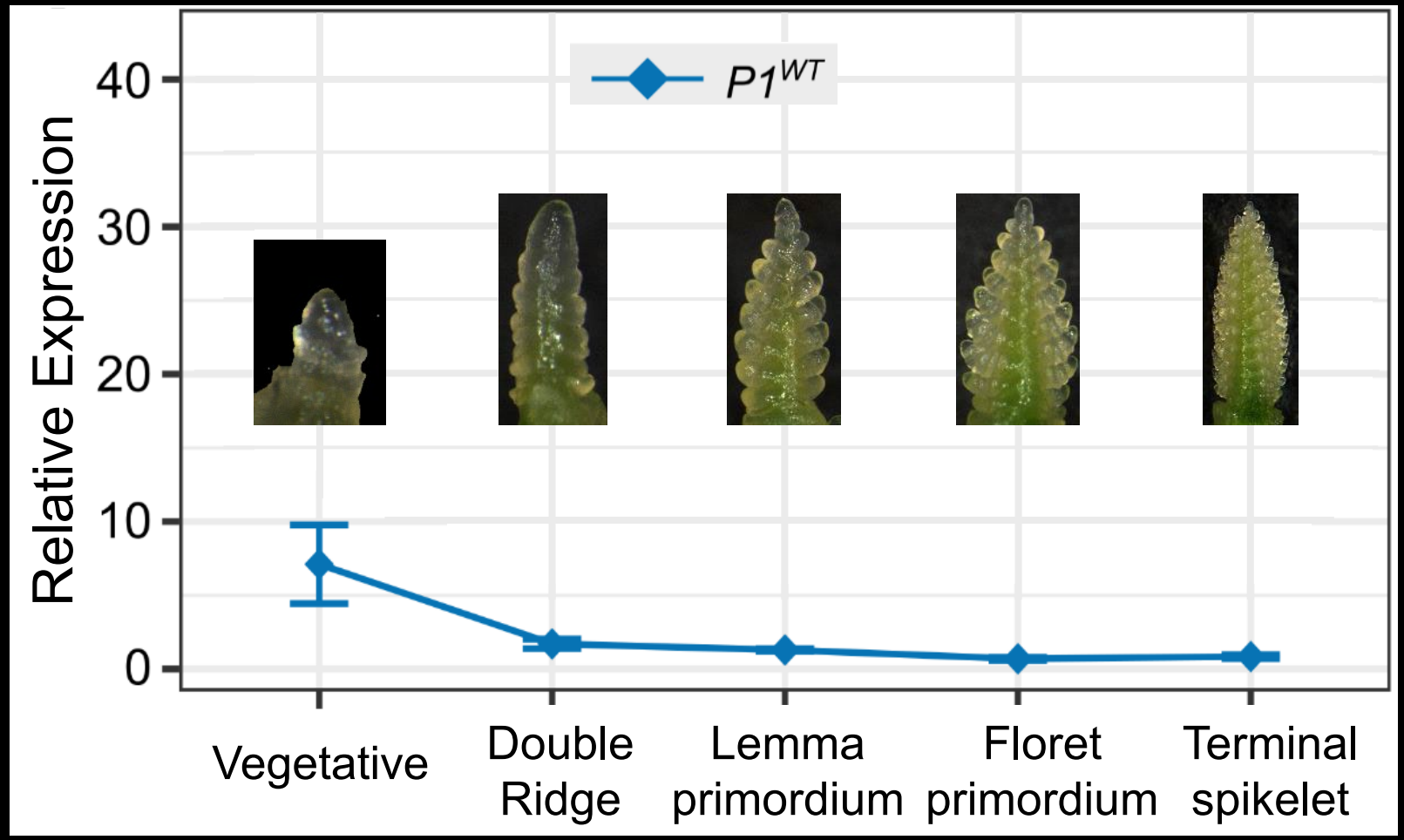


Conservation of *VRT-A2a* intron 1 sequence across grasses

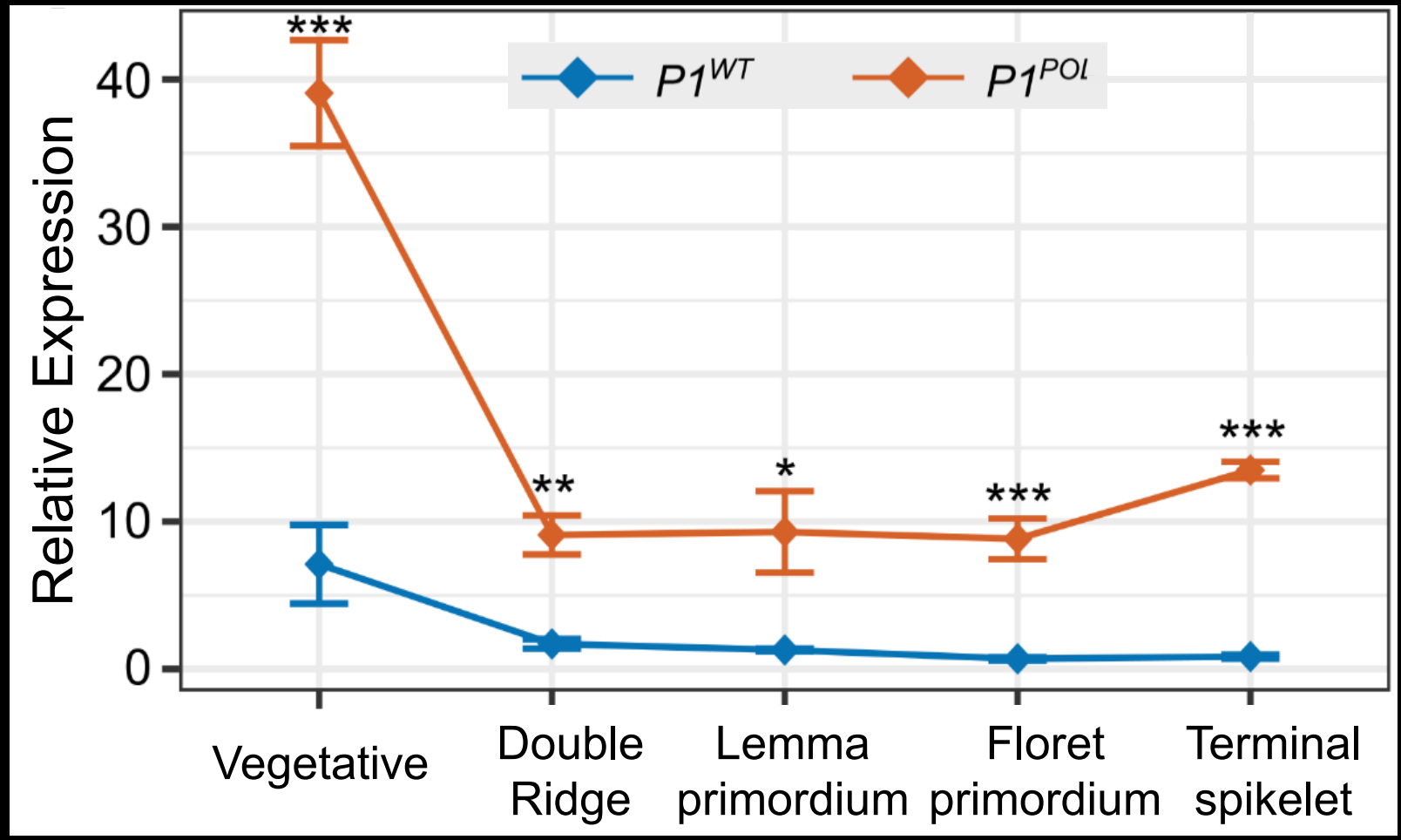


Motifs absent from *T. polonicum*

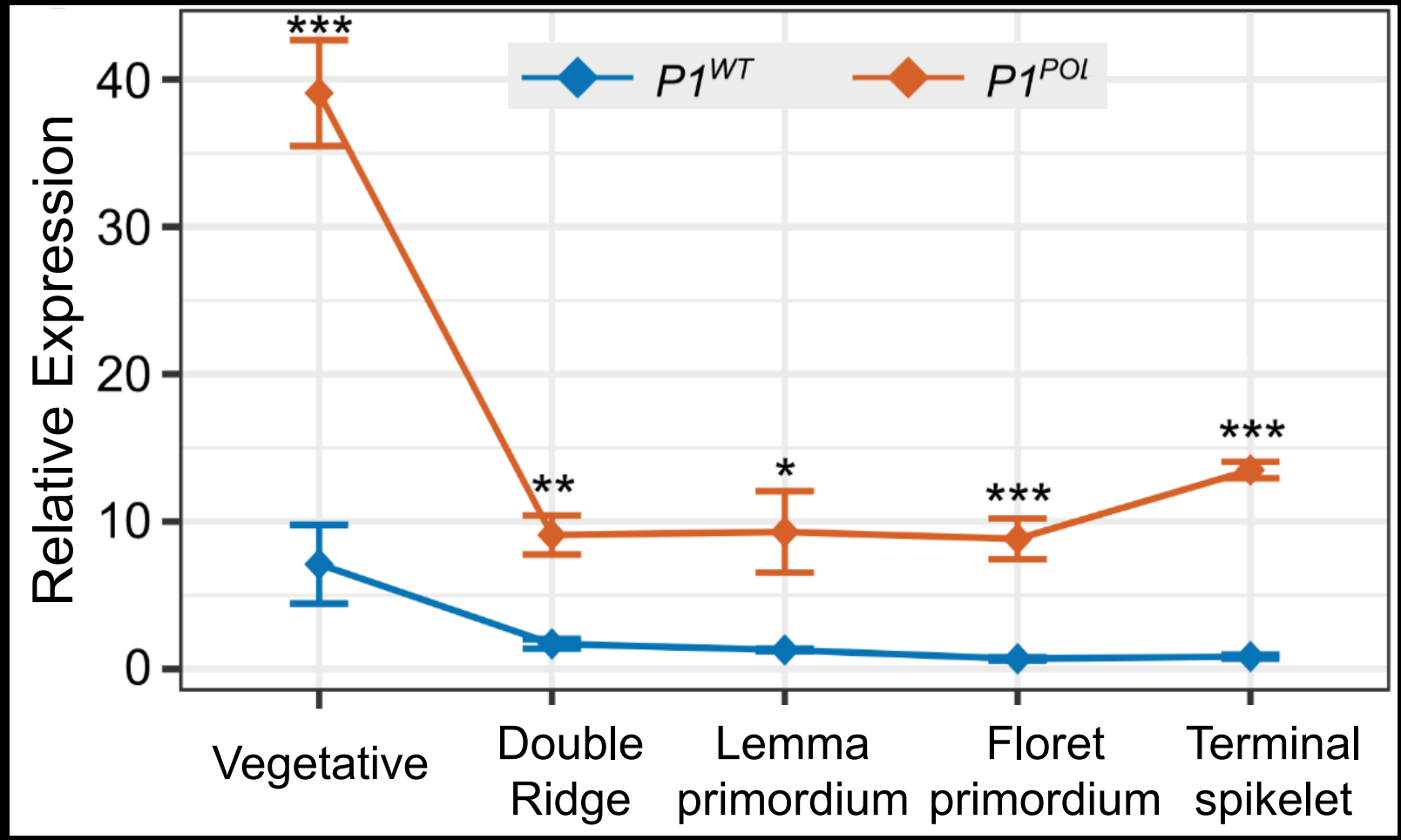
The *VRT-A2b* allele is misexpressed in developing spikes



The *VRT-A2b* allele is misexpressed in developing spikes

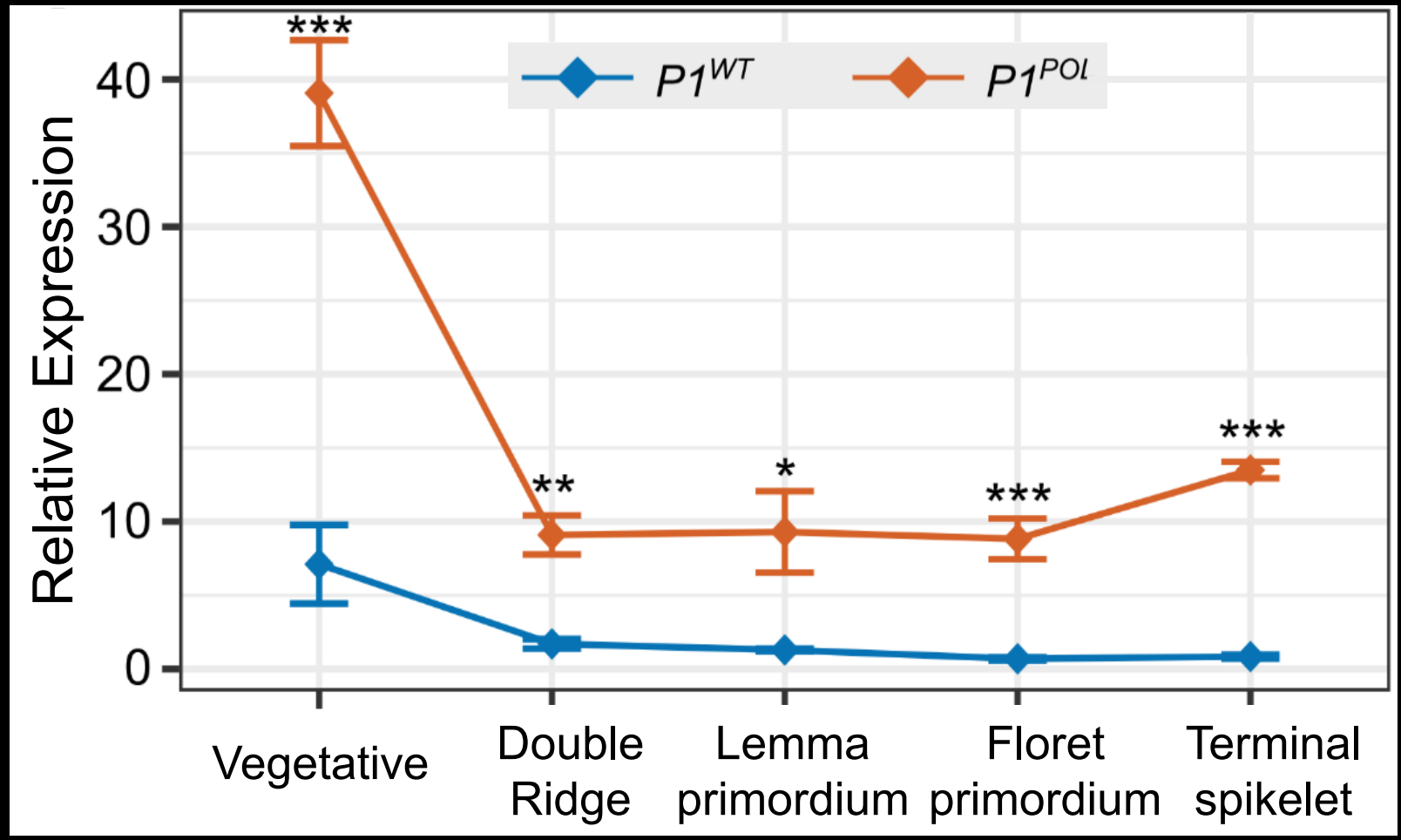


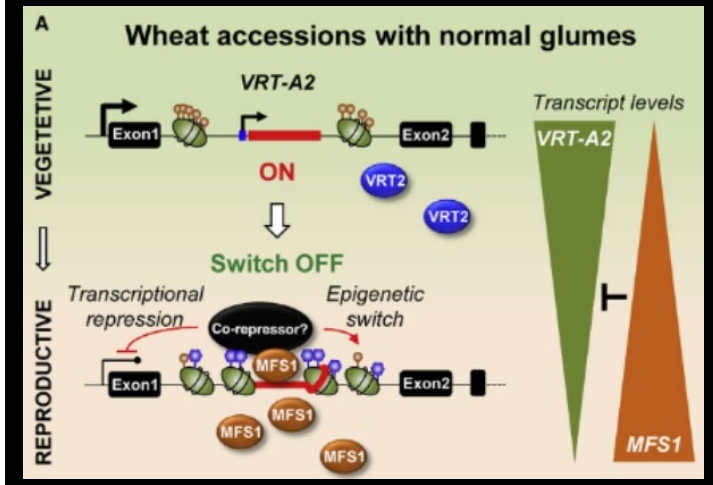
The *VRT-A2b* allele is misexpressed in developing spikes



The deleted motifs seem to act as binding sites for repressors of *VRT-A2* transcription.

The *VRT-A2b* allele is misexpressed in developing spikes

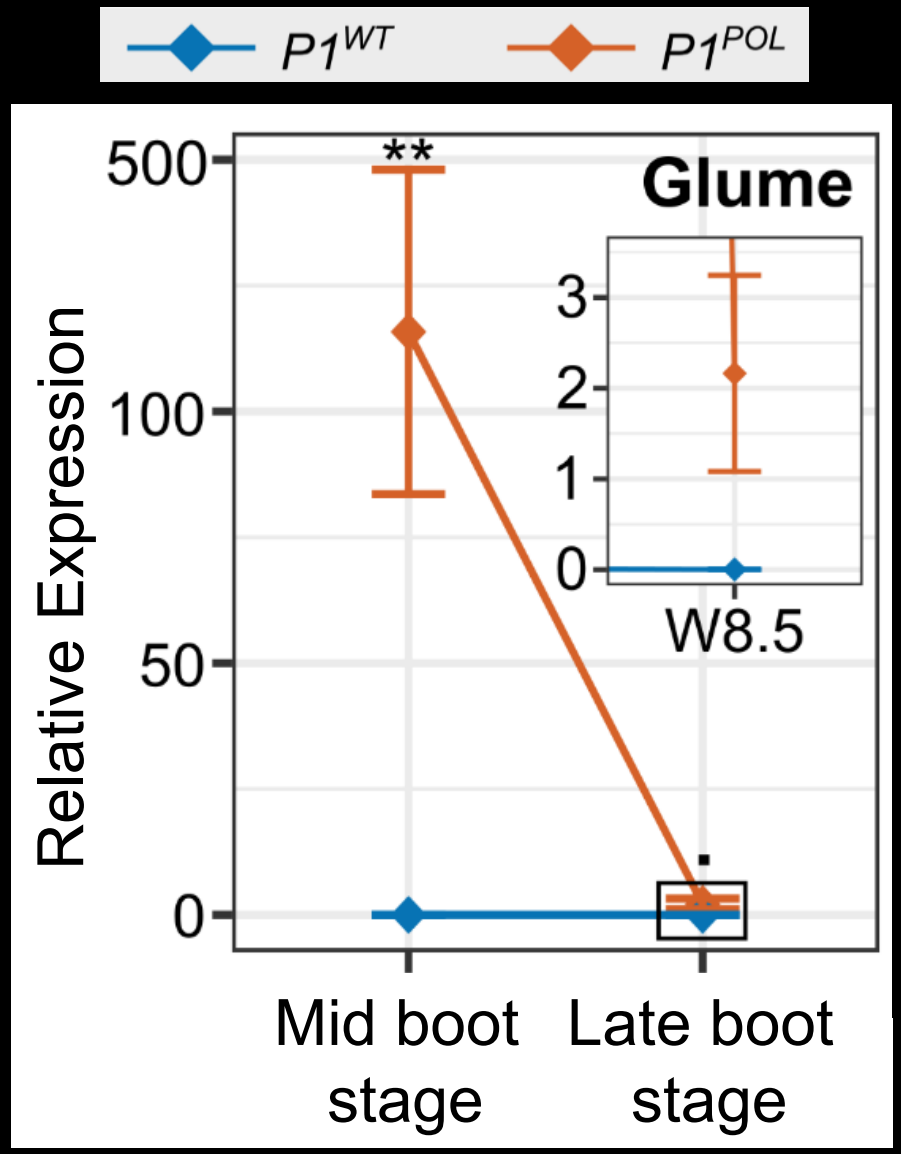




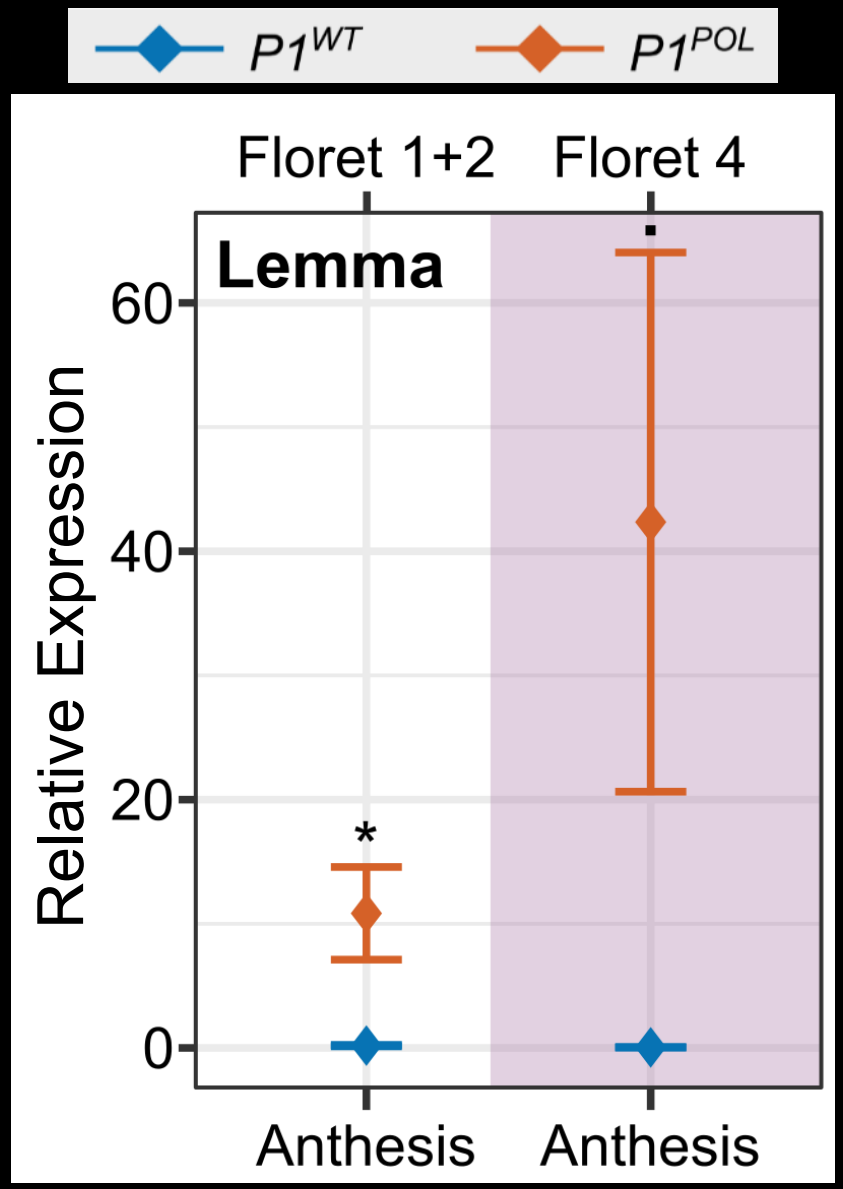
Liu *et al.*, 2021

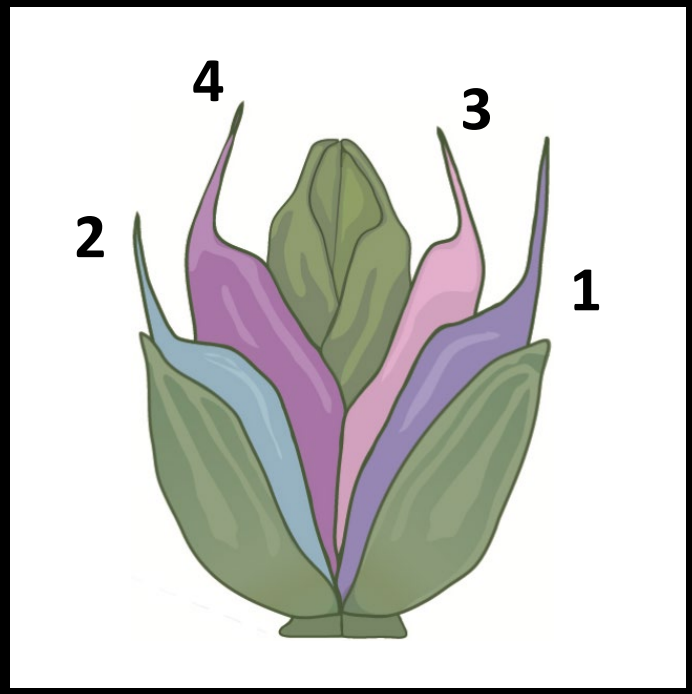
The deleted motifs seem to act as binding sites for repressors of *VRT-A2* transcription.

VRT-A2b is ectopically expressed in glumes

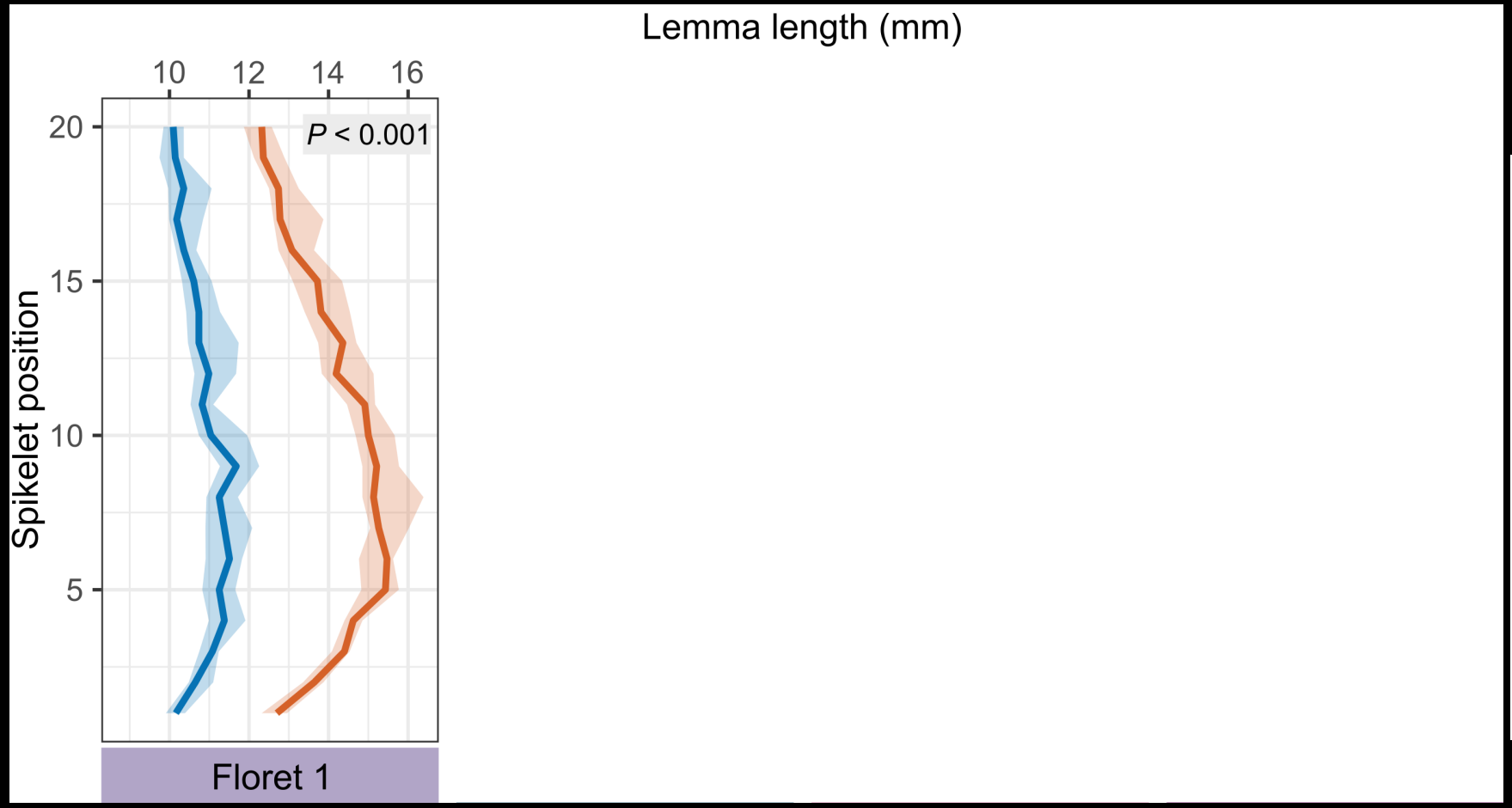


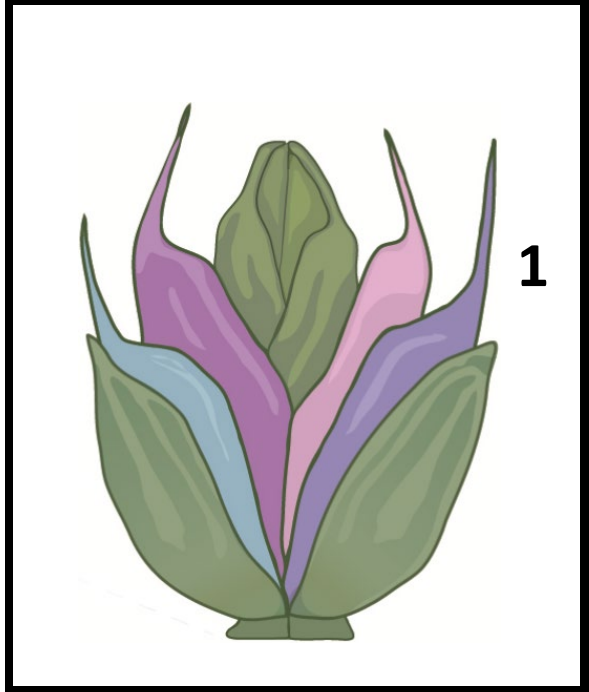
VRT-A2b is ectopically expressed in lemmas



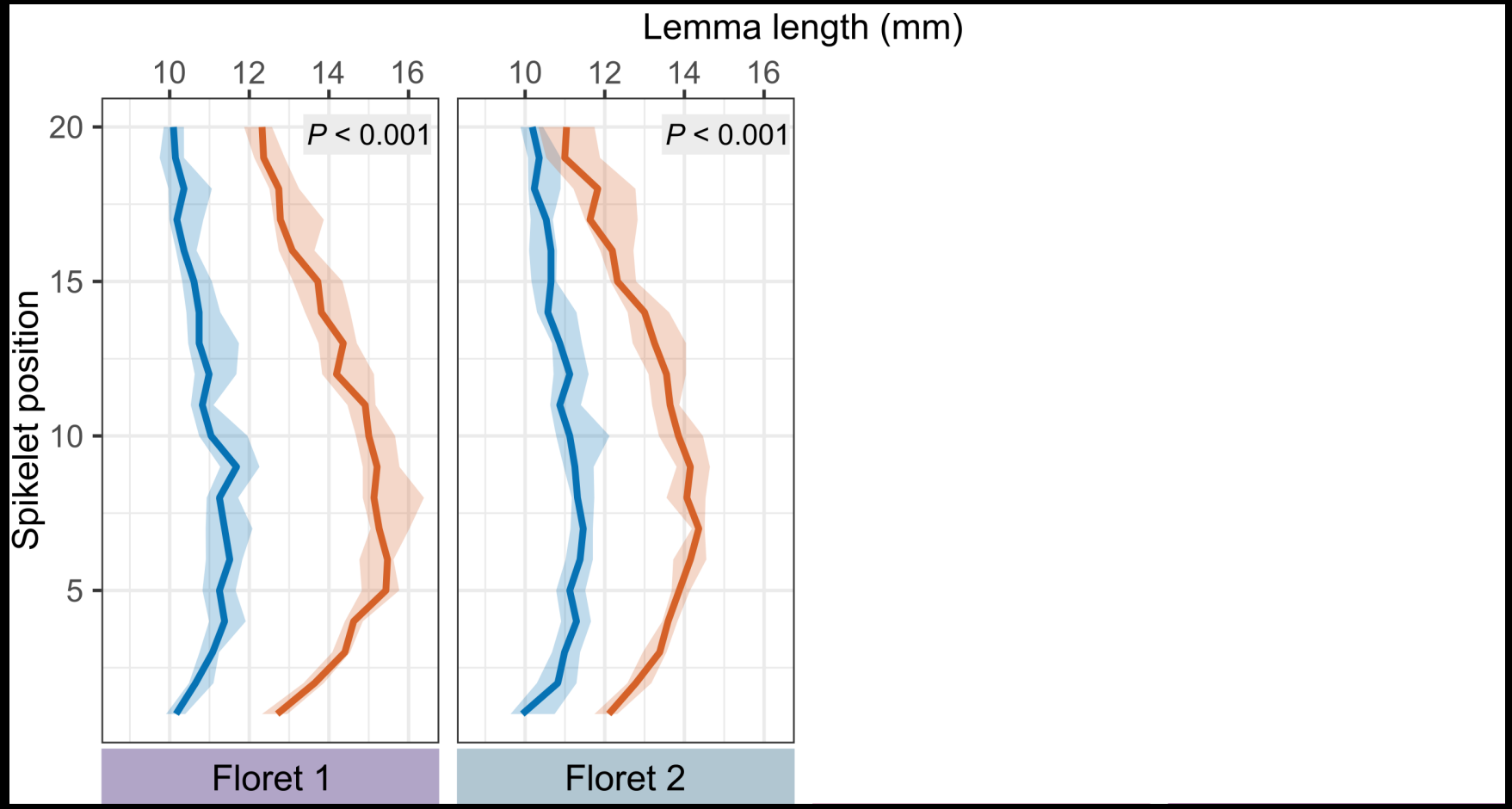


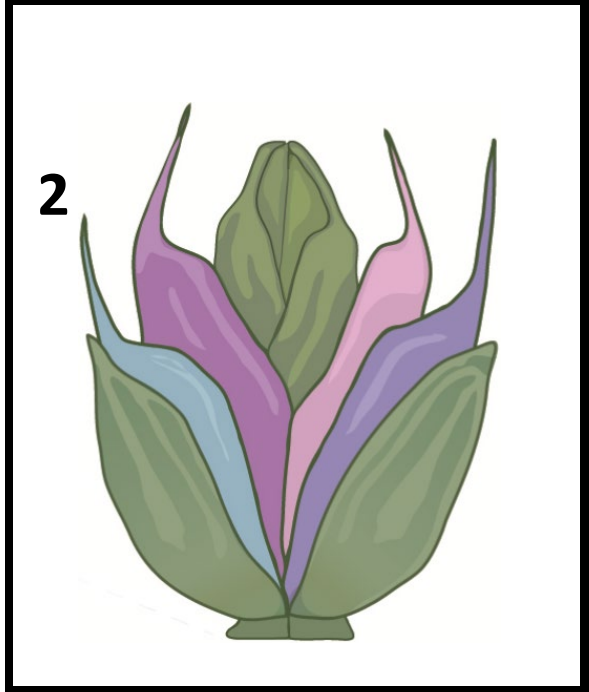
VRT-A2b affects outer and basal organs of spikelets



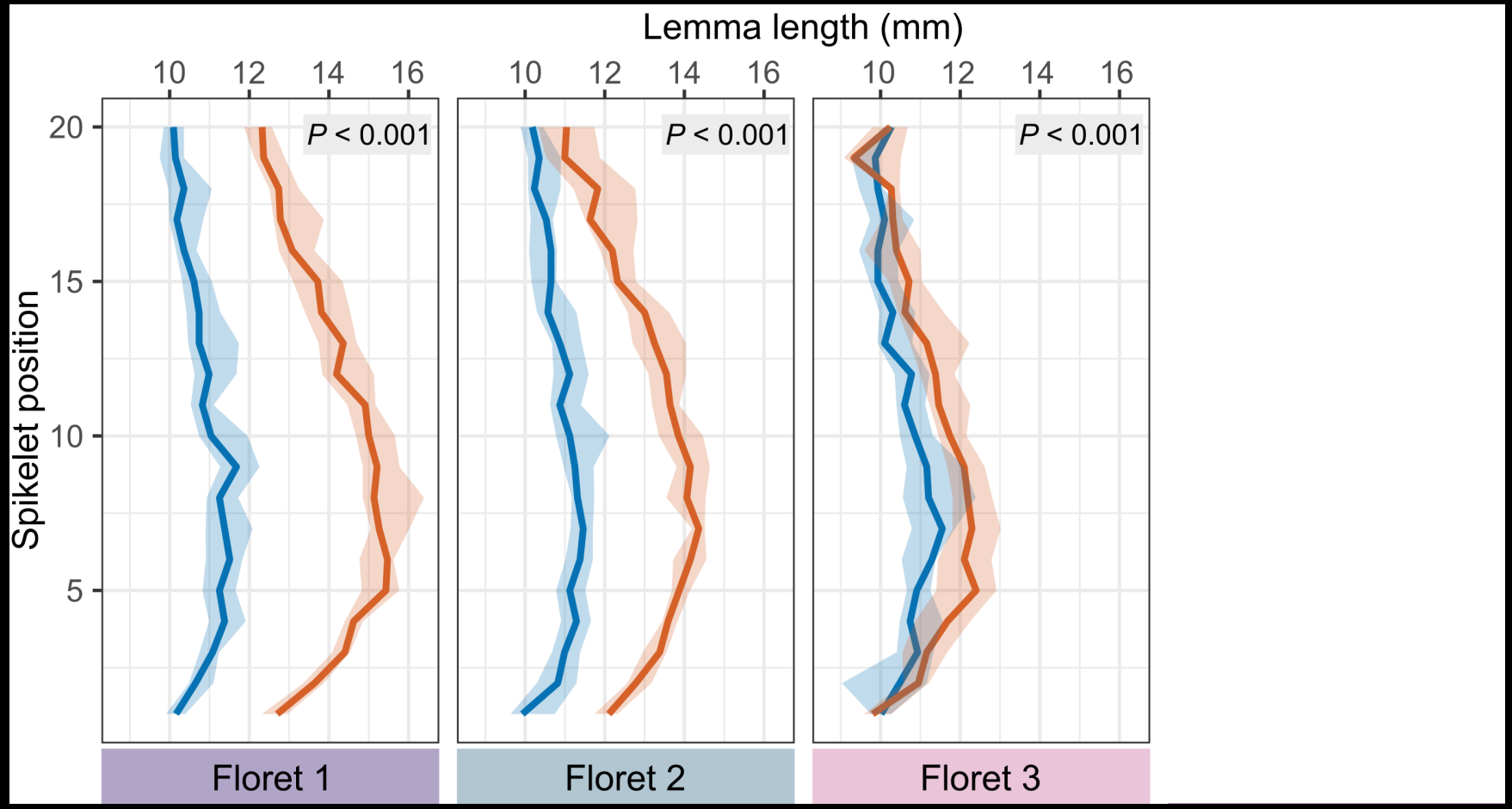


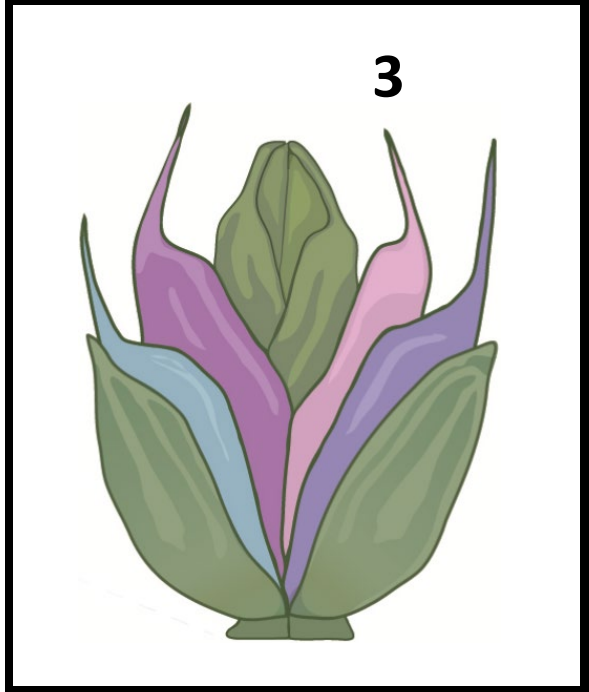
VRT-A2b affects outer and basal organs of spikelets





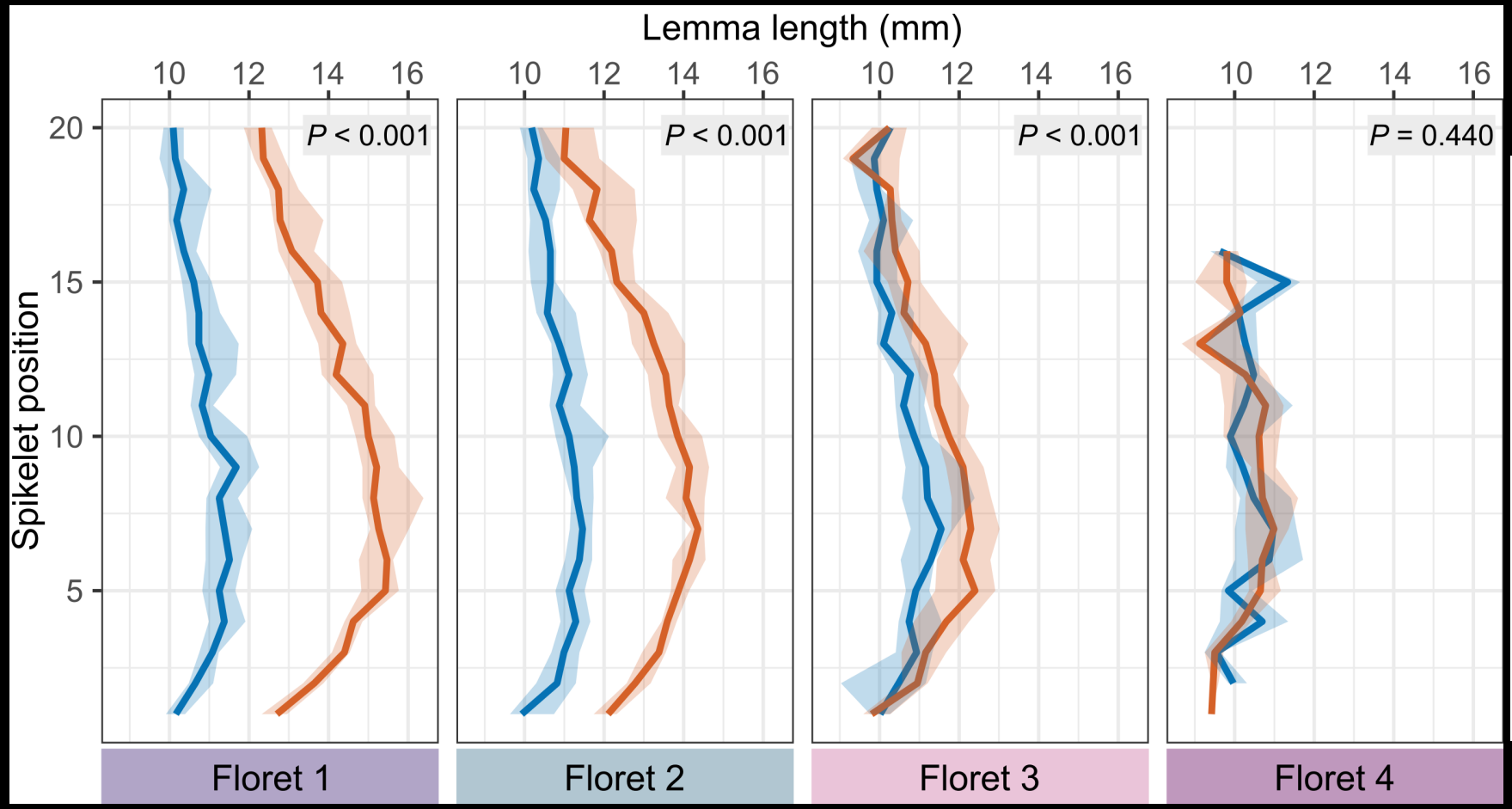
VRT-A2b affects outer and basal organs of spikelets

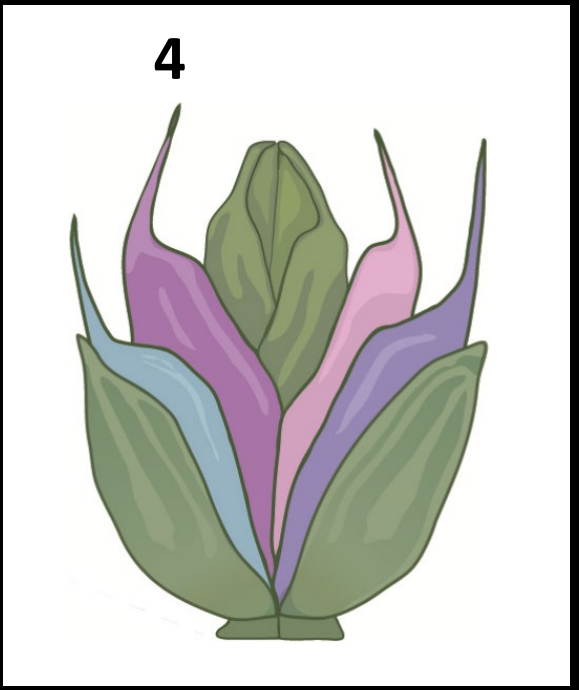




$P1^{WT}$ $P1^{POL}$

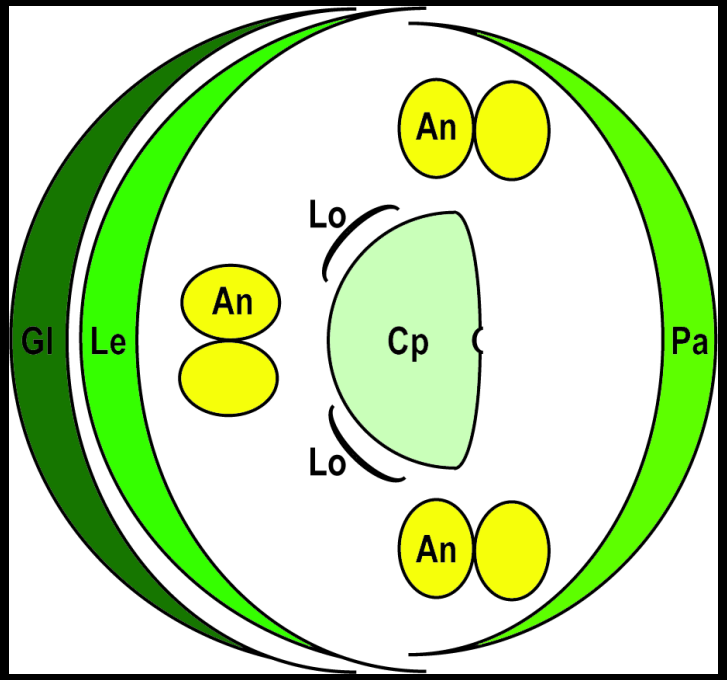
VRT-A2b affects outer and basal organs of spikelets





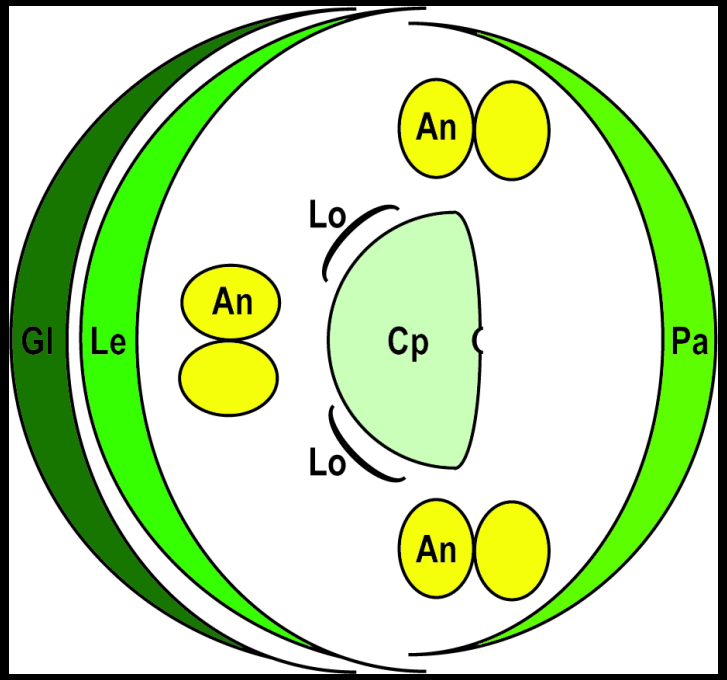
$P1^{WT}$ $P1^{POL}$

VRT-A2b affects outer and basal organs of spikelets



Cross-section

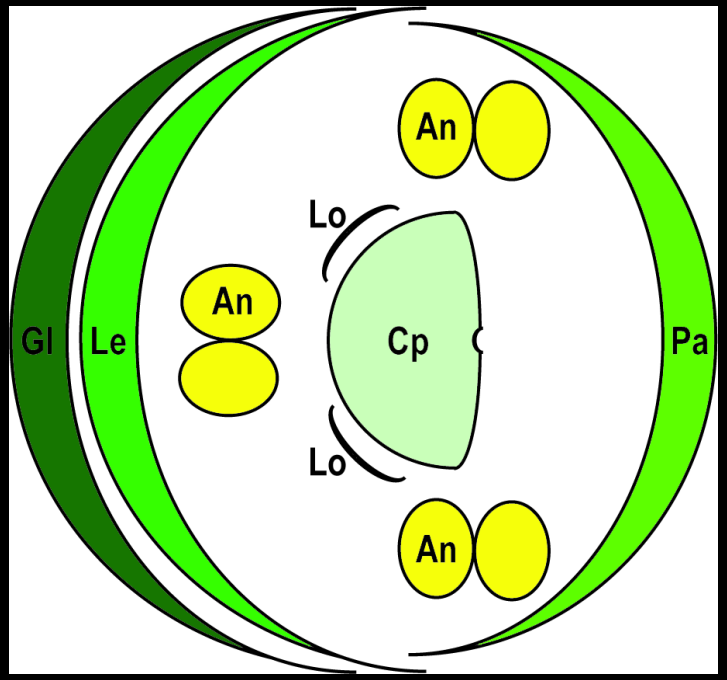
VRT-A2b affects outer and basal organs of spikelets



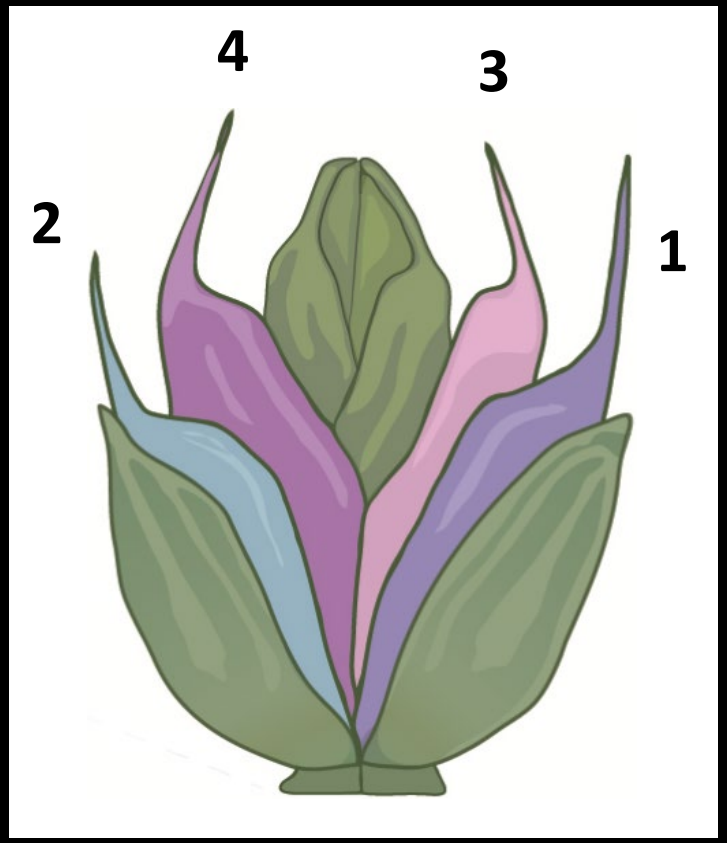
Outer Inner

Cross-section

VRT-A2b affects outer and basal organs of spikelets



Outer Inner

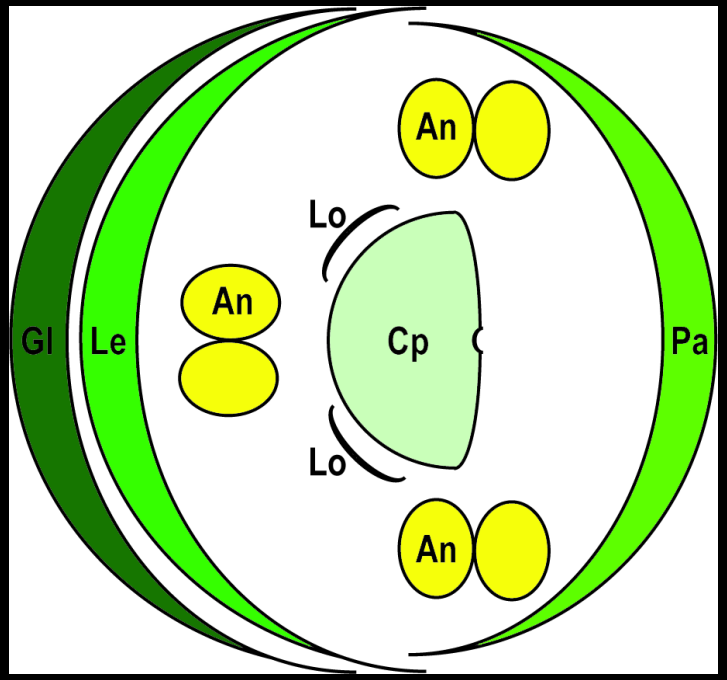


Floret 4
Floret 3
Floret 2
Floret 1

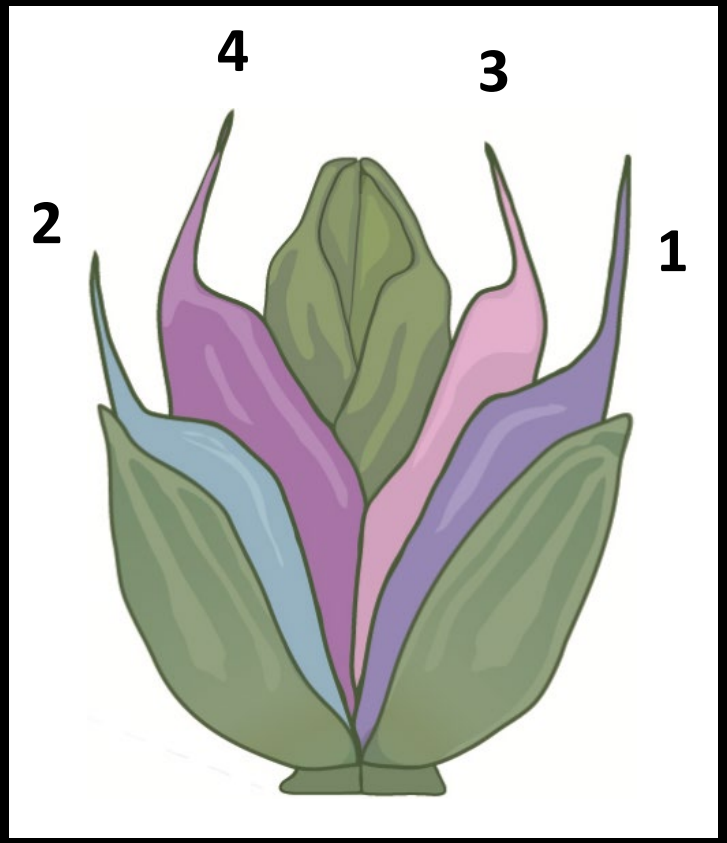
Apical

Basal

VRT-A2b affects outer and basal organs of spikelets



Older → Younger

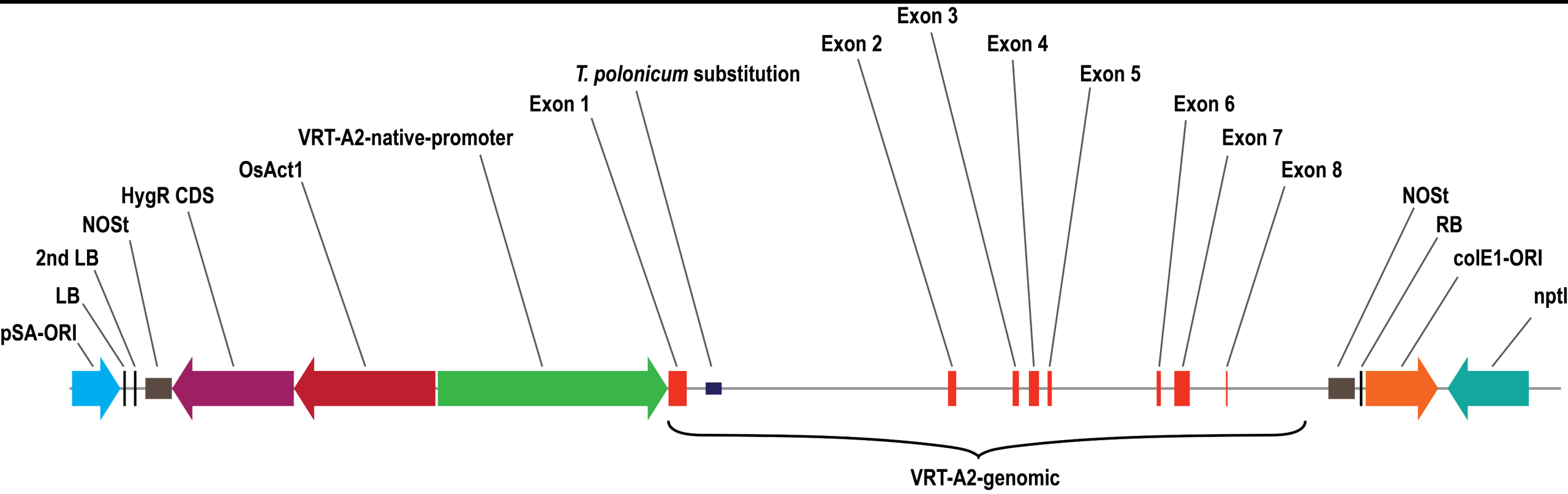


Floret 4
Floret 3
Floret 2
Floret 1

Younger

Older

Complementation of “Fielder” with *VRT-A2b* allele

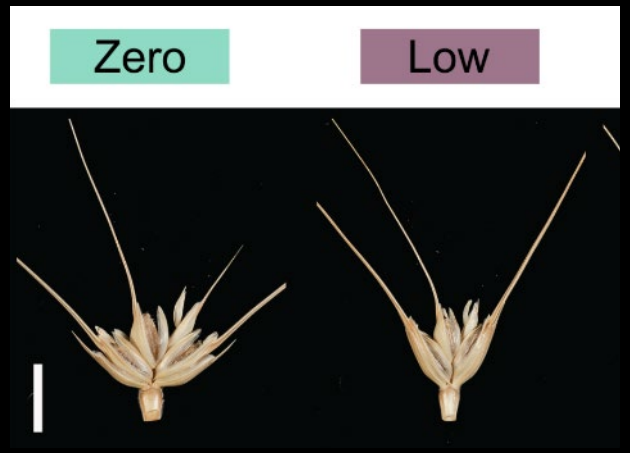


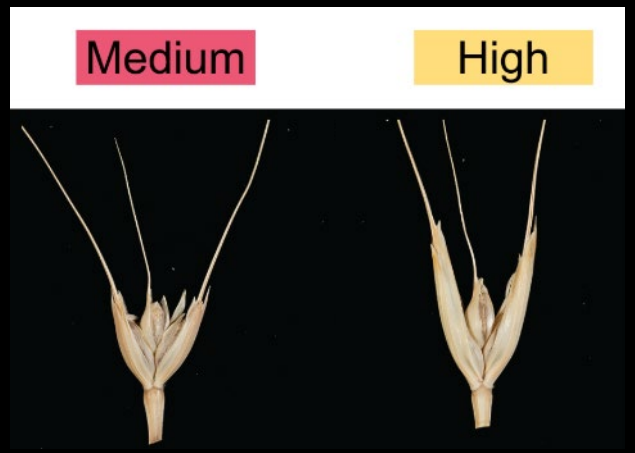
Sadiye Hayta

Mark Smedley

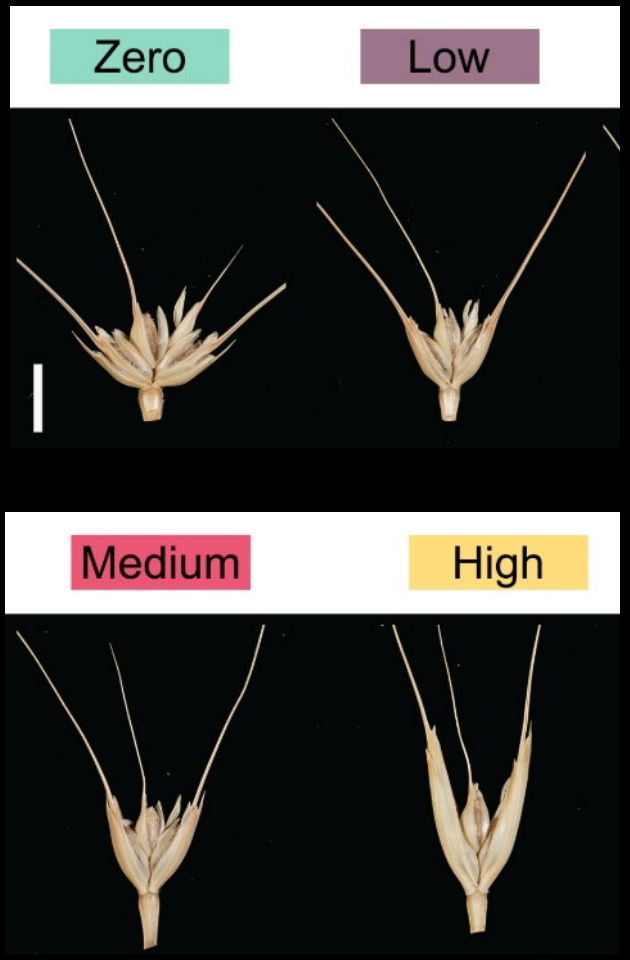
Hayta *et al.*, 2019

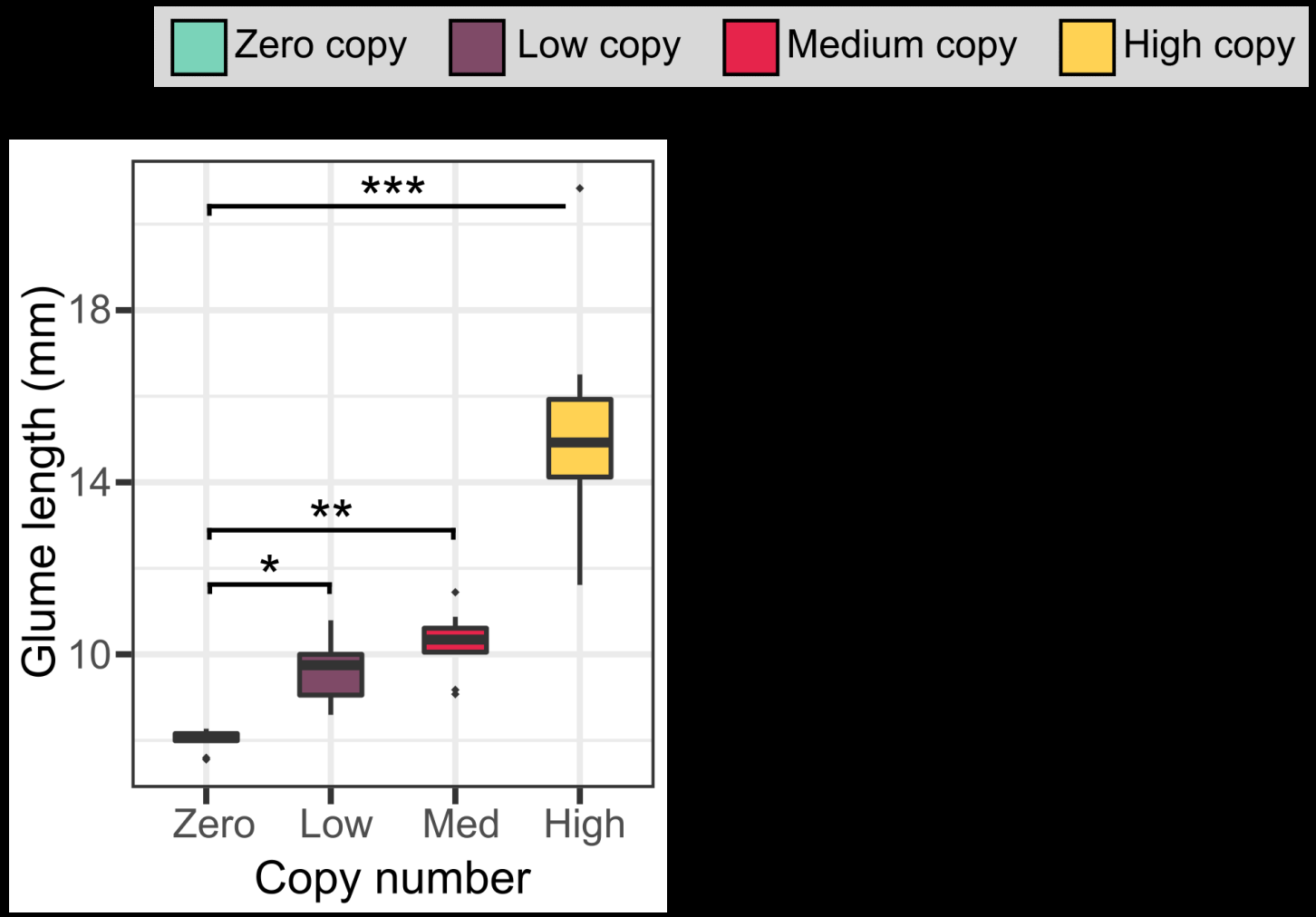
VRT-A2b acts in a dosage-dependent manner in T1 lines



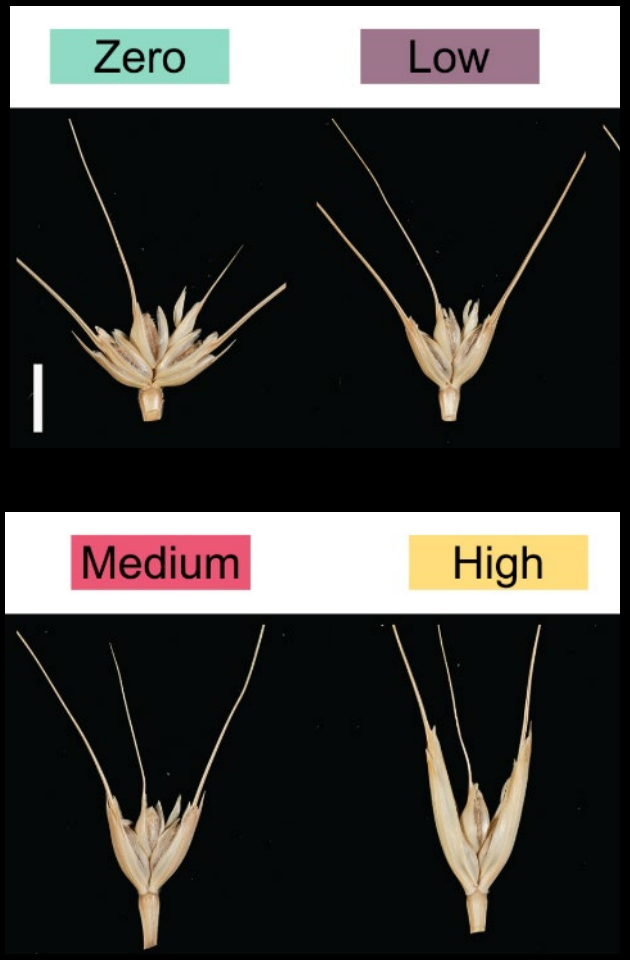


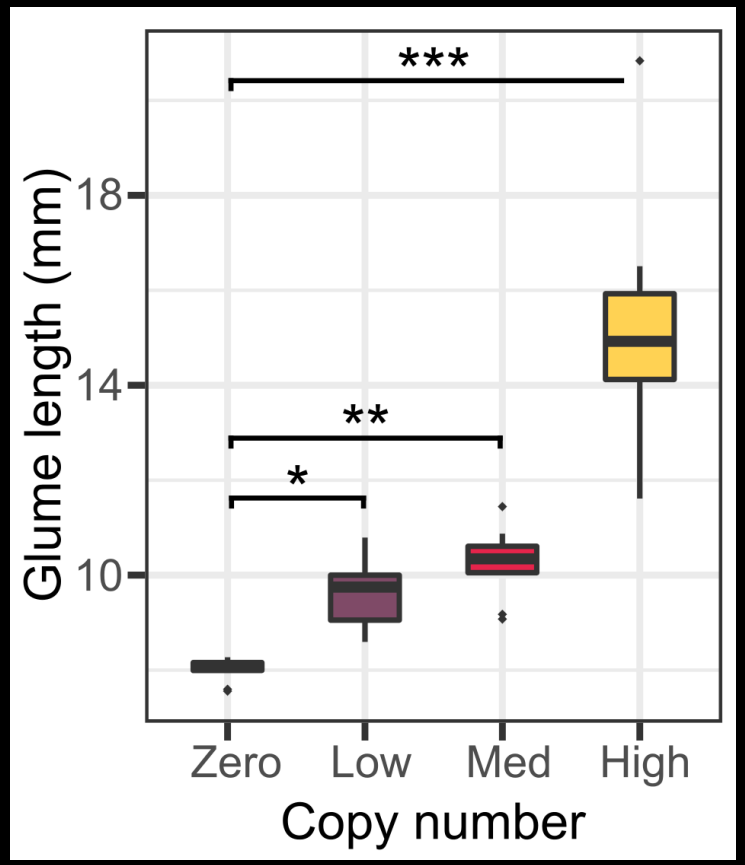
VRT-A2b acts in a dosage-dependent manner in T1 lines

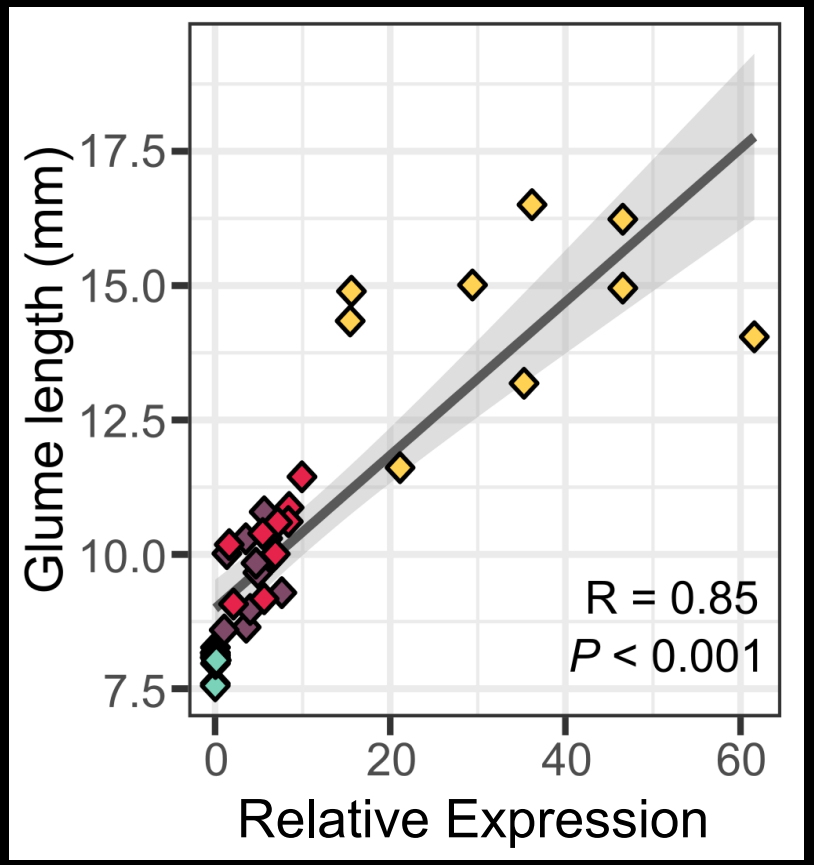




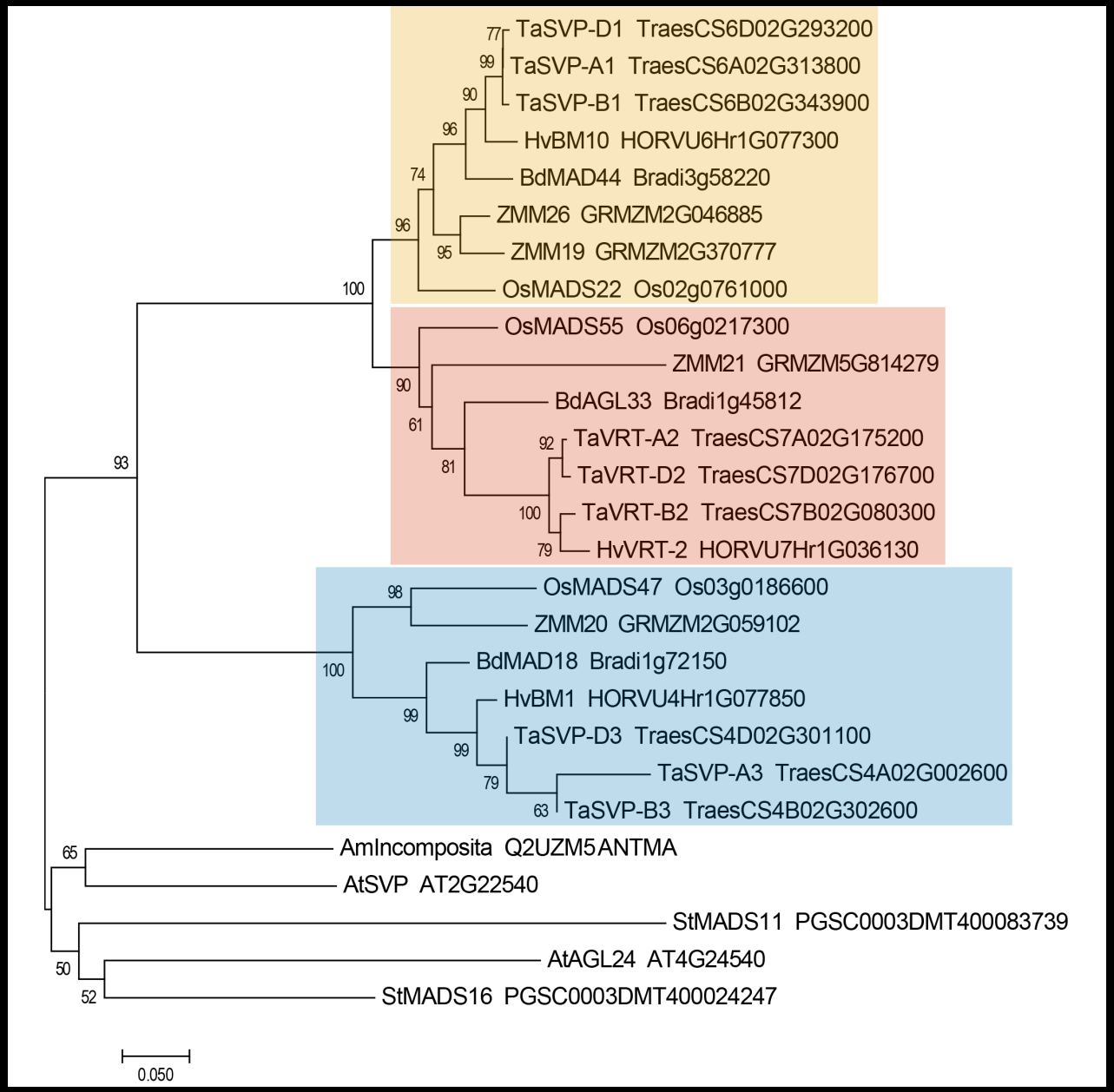
VRT-A2b acts in a dosage-dependent manner in T1 lines





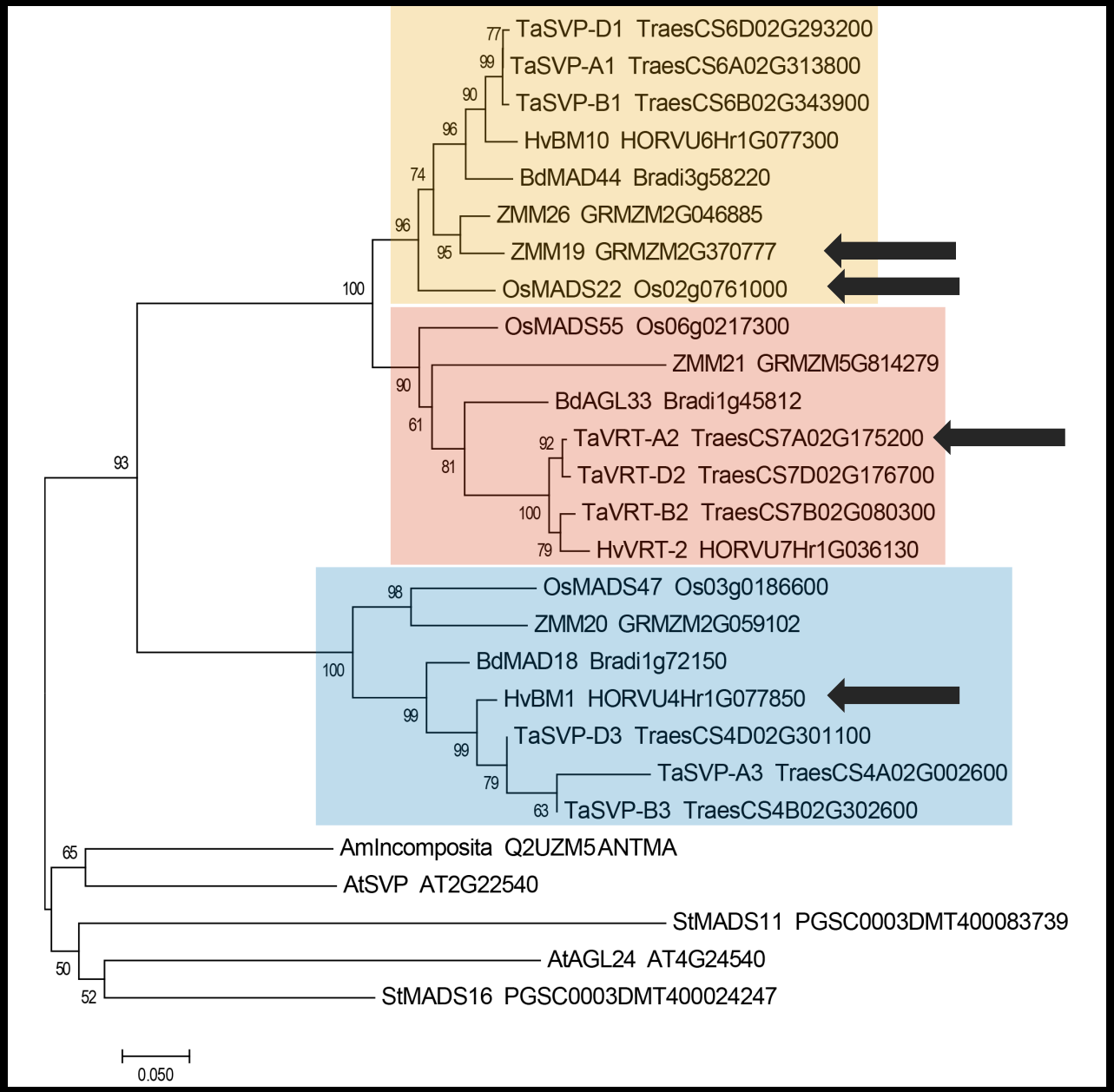


SVP family in grasses



Triplication of *SVP* genes in grasses

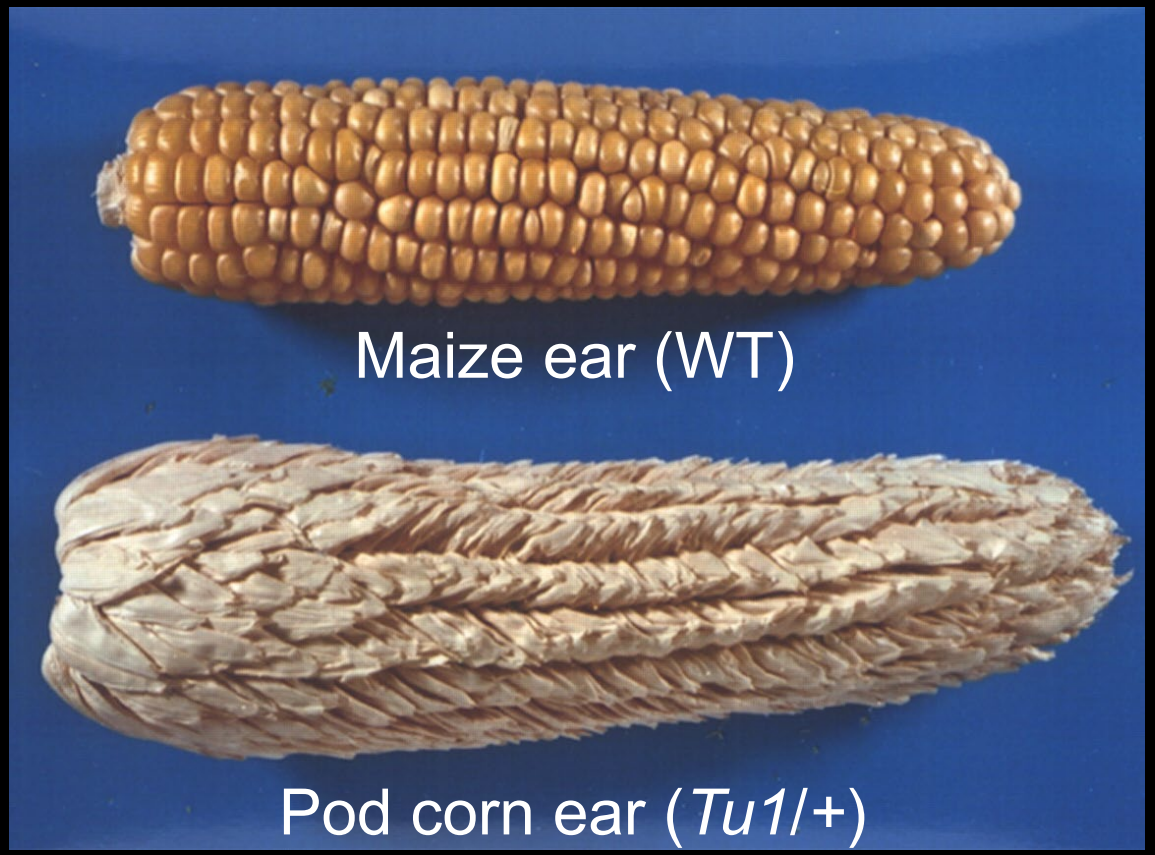
SVP family in grasses



SVP members shown to cause elongated glume or lemma.

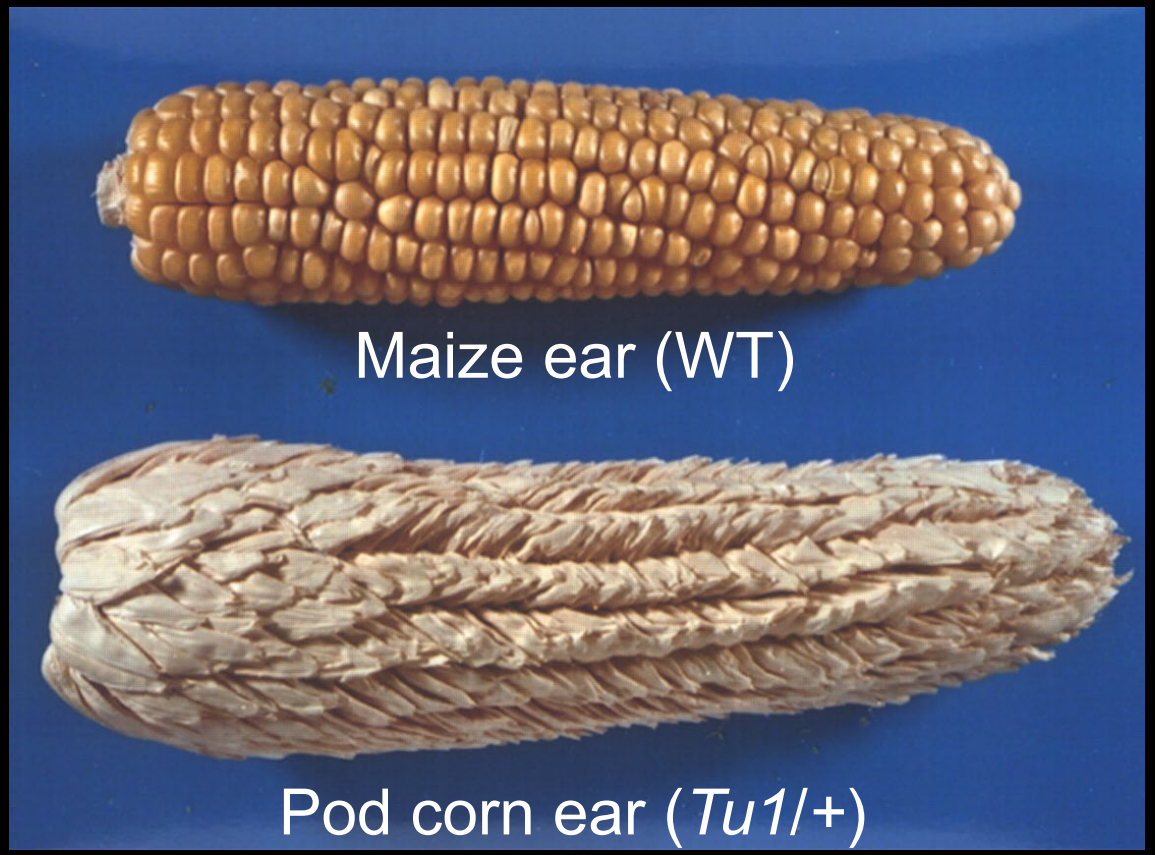
Sentoku et al., 2005; Trevaskis *et al.,* 2007; Wingen *et al.,* 2012; Han *et al.,* 2012

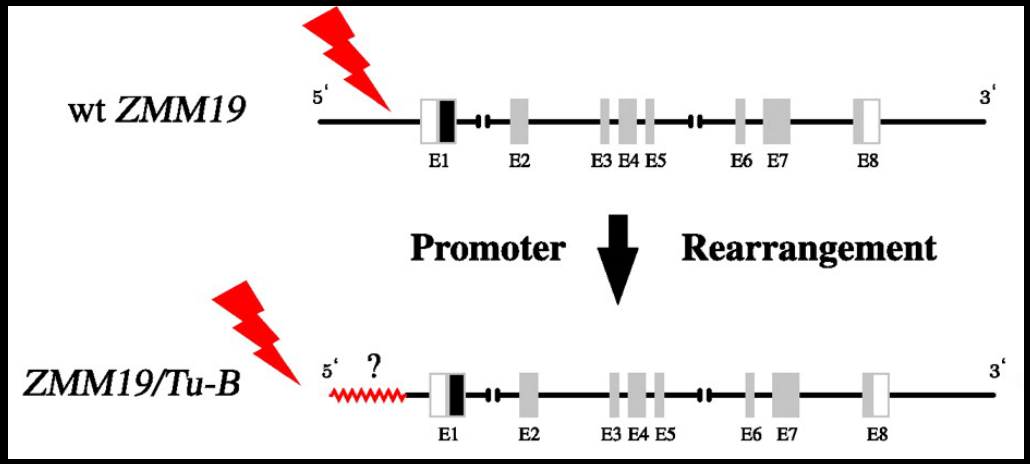
VRT-A2b reminiscent of “pod corn” allele in maize



Wingen *et al.*, 2012; Han *et al.*, 2012

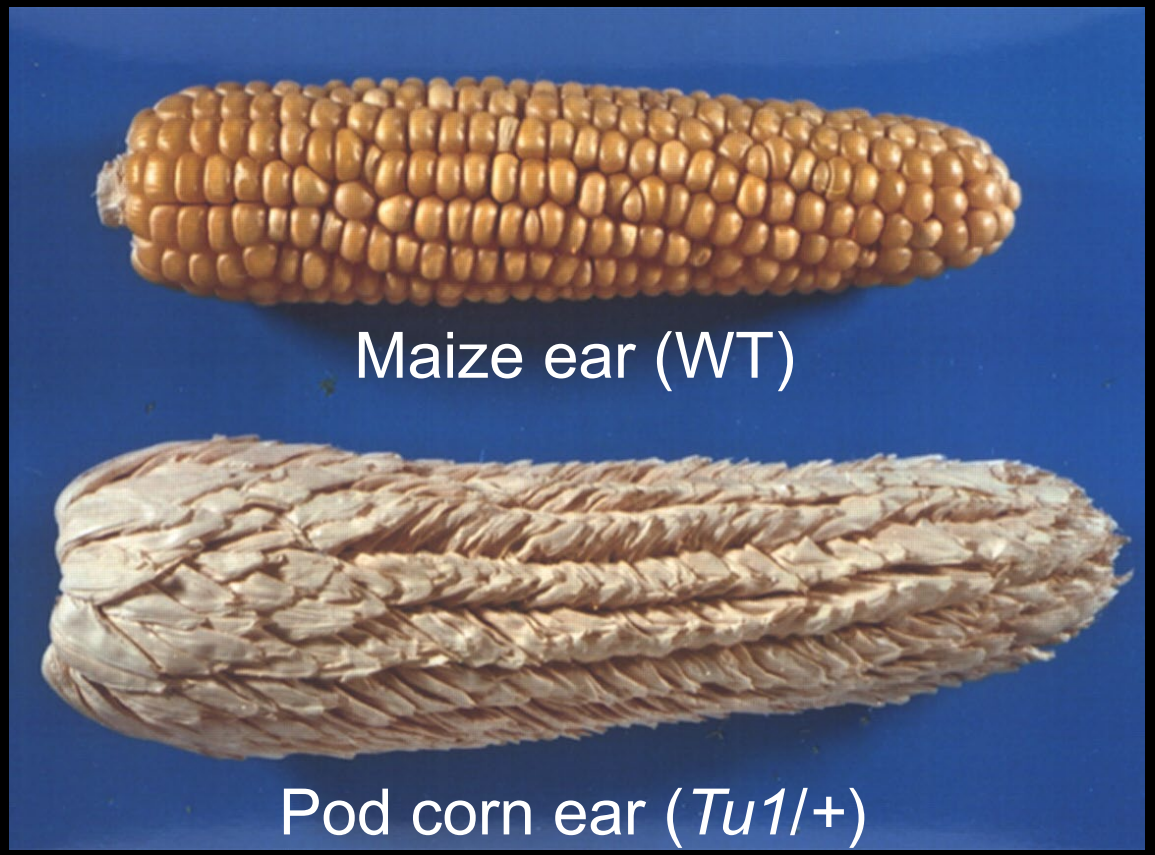
VRT-A2b reminiscent of “pod corn” allele in maize

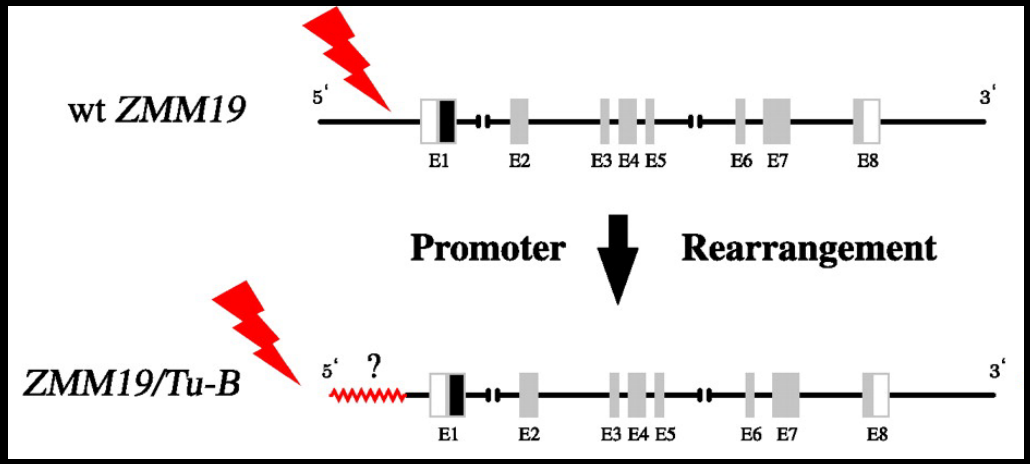


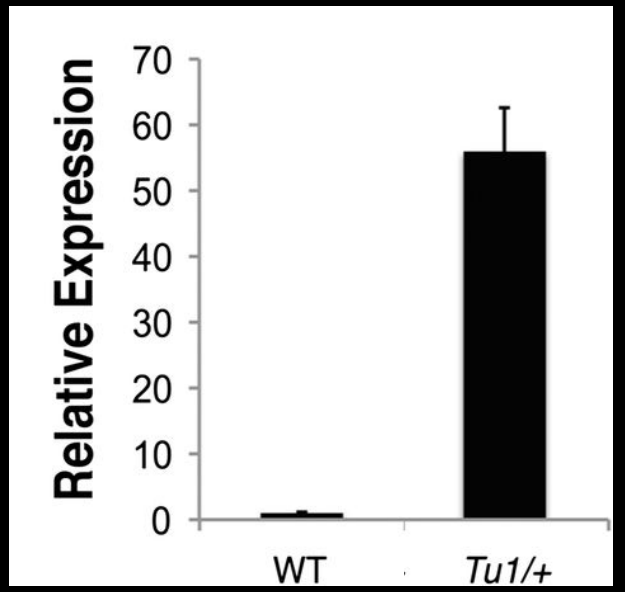


Wingen *et al.*, 2012; Han *et al.*, 2012

VRT-A2b reminiscent of "pod corn" allele in maize

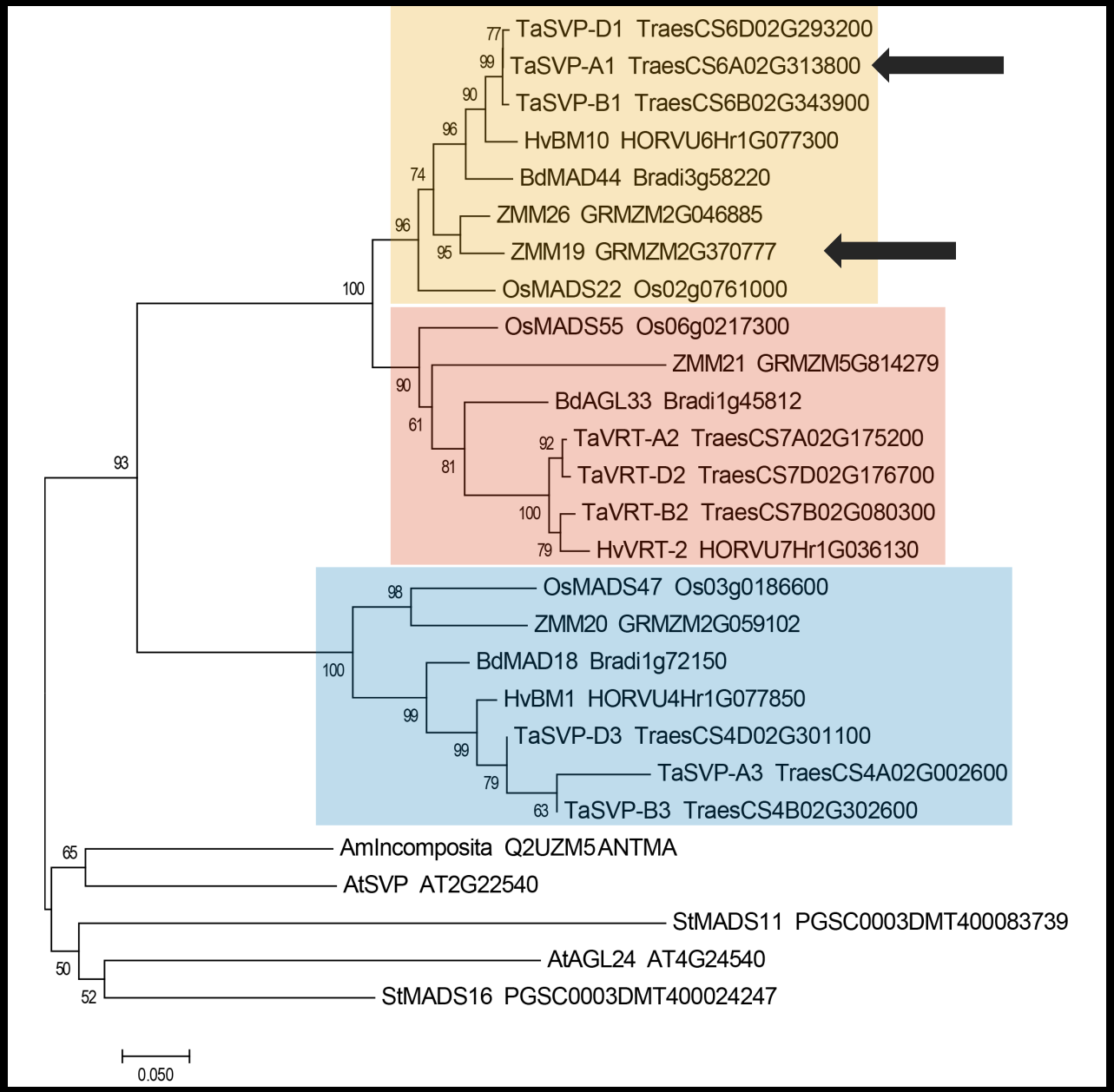






Wingen *et al.*, 2012; Han *et al.*, 2012

SVP family in grasses

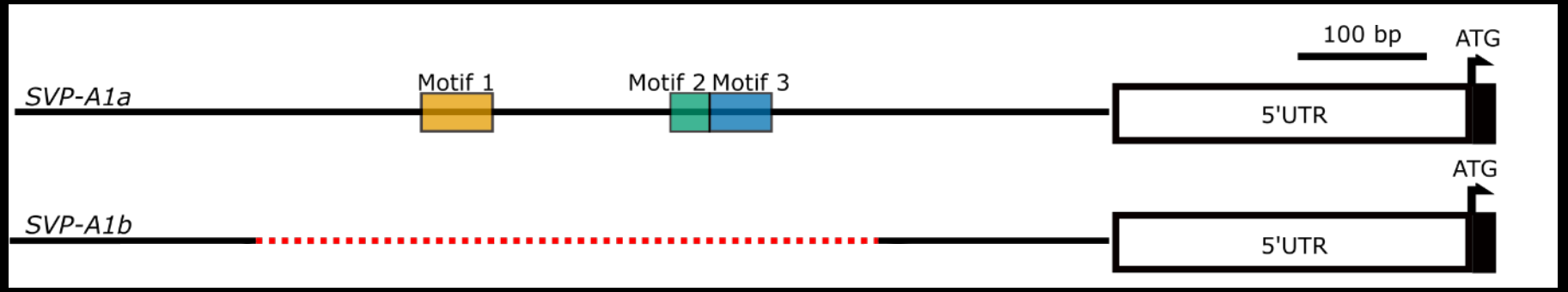


TaSVP-A1 linked to long glumes in *T. ispahanicum*

Yi (Andy) Chen

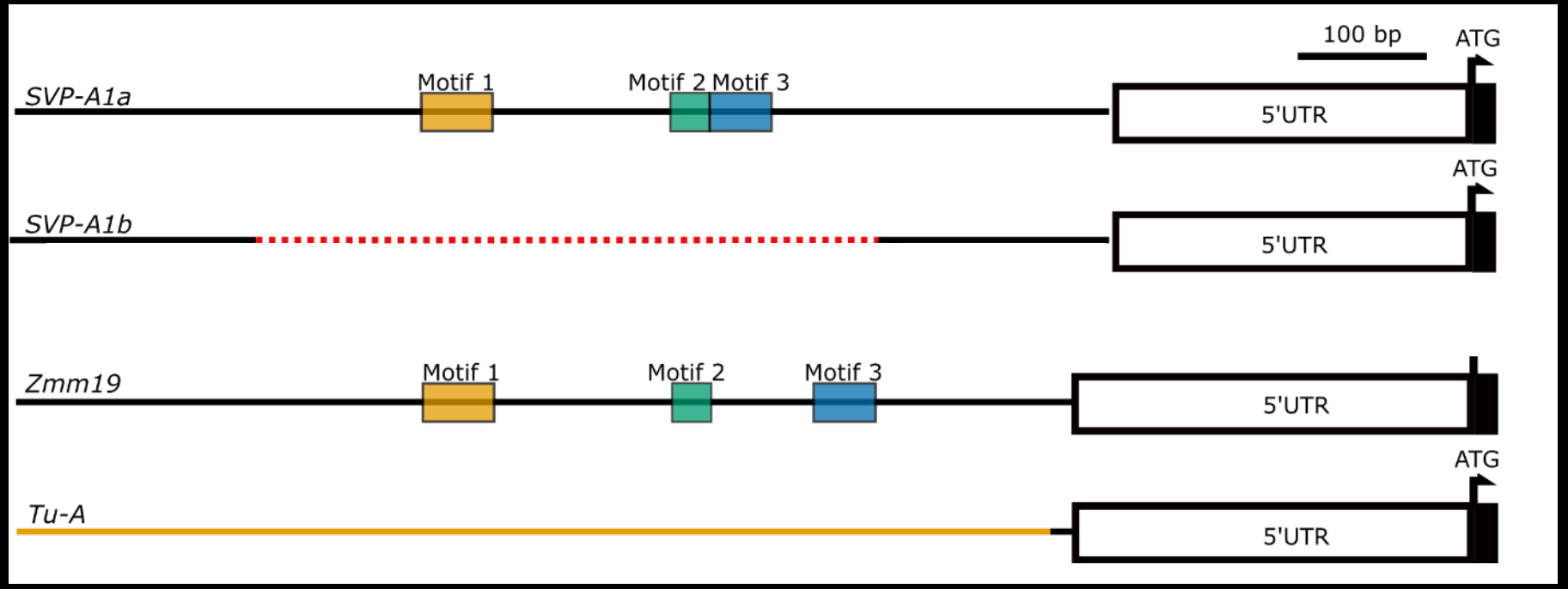
Chen *et al.*, 2022; https://doi.org/10.1101/2022.01.27.478079

TaSVP-A1 linked to long glumes in *T. ispahanicum*



Chen *et al.*, 2022; https://doi.org/10.1101/2022.01.27.478079

TaSVP-A1 linked to long glumes in *T. ispahanicum*



Chen *et al.*, 2022; https://doi.org/10.1101/2022.01.27.478079

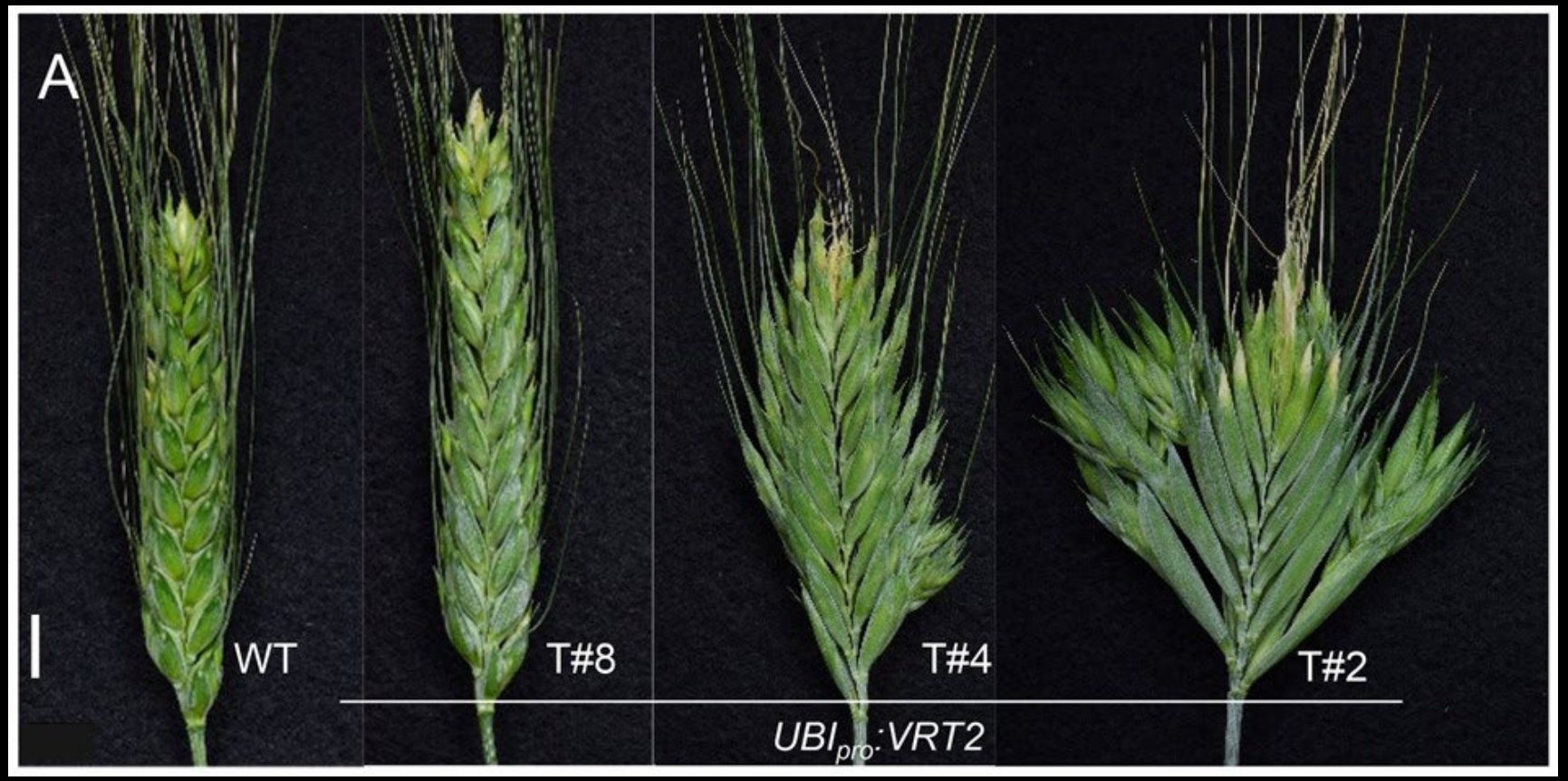
Transcription levels of *SVP*s essential for spike development

WT UBI::HvSVP1

Trevaskis *et al.*, 2007

Transcription levels of *SVP*s essential for spike development

WT UBI::HvSVP1



VRT2 transcription levels

Trevaskis *et al.,* 2007; Li *et al.,* 2021

Summary

- *SVP*s play a crucial role in grass spike development

- (Core) Functional regulation (motifs) conserved between distantly related grasses

- *SVP*s act in a dosage-dependent manner

- Modulating the expression pattern of *SVP*s holds potential to improve agronomic traits

Next steps

- Investigate motifs in intron 1 of *VRT-A2*
    - Is loss of one or both motifs necessary for "*polonicum*" phenotype?
    - Which proteins bind the intron 1 motifs?
- Elucidate genetic network surrounding *VRT2*
    - Upstream regulators
    - Downstream targets

Thank you for listening

James Simmonds
Jemima Brinton
Anna Backhaus
Andy Chen
Mark Smedley
Sadiye Hayta
Pamela Crane
Tobin Florio
Elaine Barclay
Cristobal Uauy

Adamski *et al.*, 2021
https://doi.org/10.1093/plcell/koab119

@NikolaiAdamski

John Innes Centre

UKRI Biotechnology and Biological Sciences Research Council

IWGSC

GRU SeedStor
IPK Genebank
USDA NSGC
CGN Wageningen