The First Step to Tackling the FN Problem:
Identifying PHS Tolerant Genes/QTL in PNW Germplasm

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PNW Quality Council

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github.com/shantel-martinez/FNWorkshop2019

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Acknowledgments

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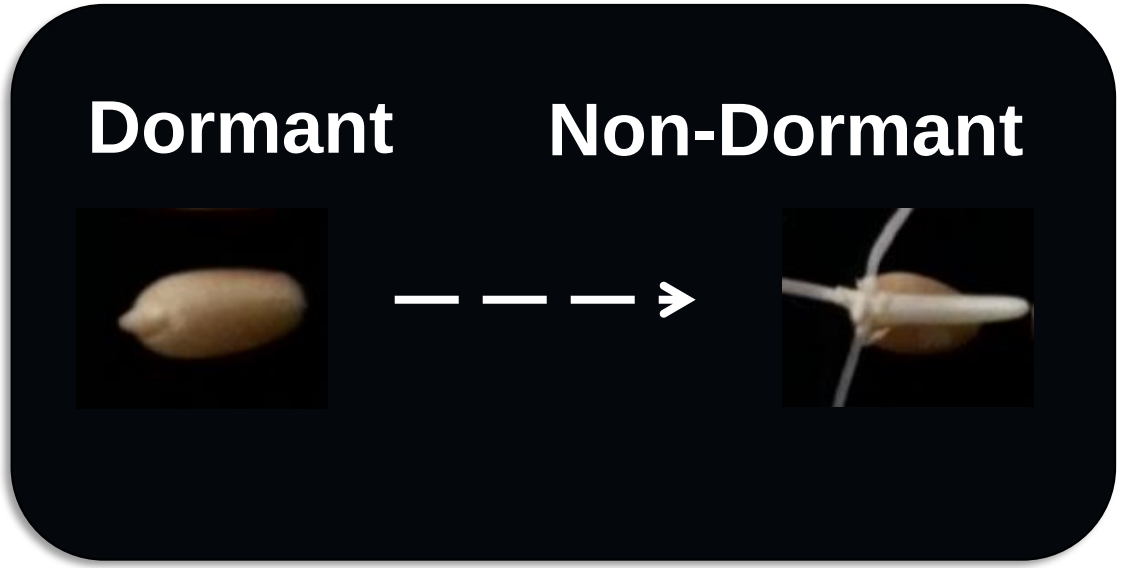
Preharvest Sprouting

Germination of mature seed on the mother plant when cool and wet conditions occur before harvest

PHS Tolerant ⟷ PHS Susceptible

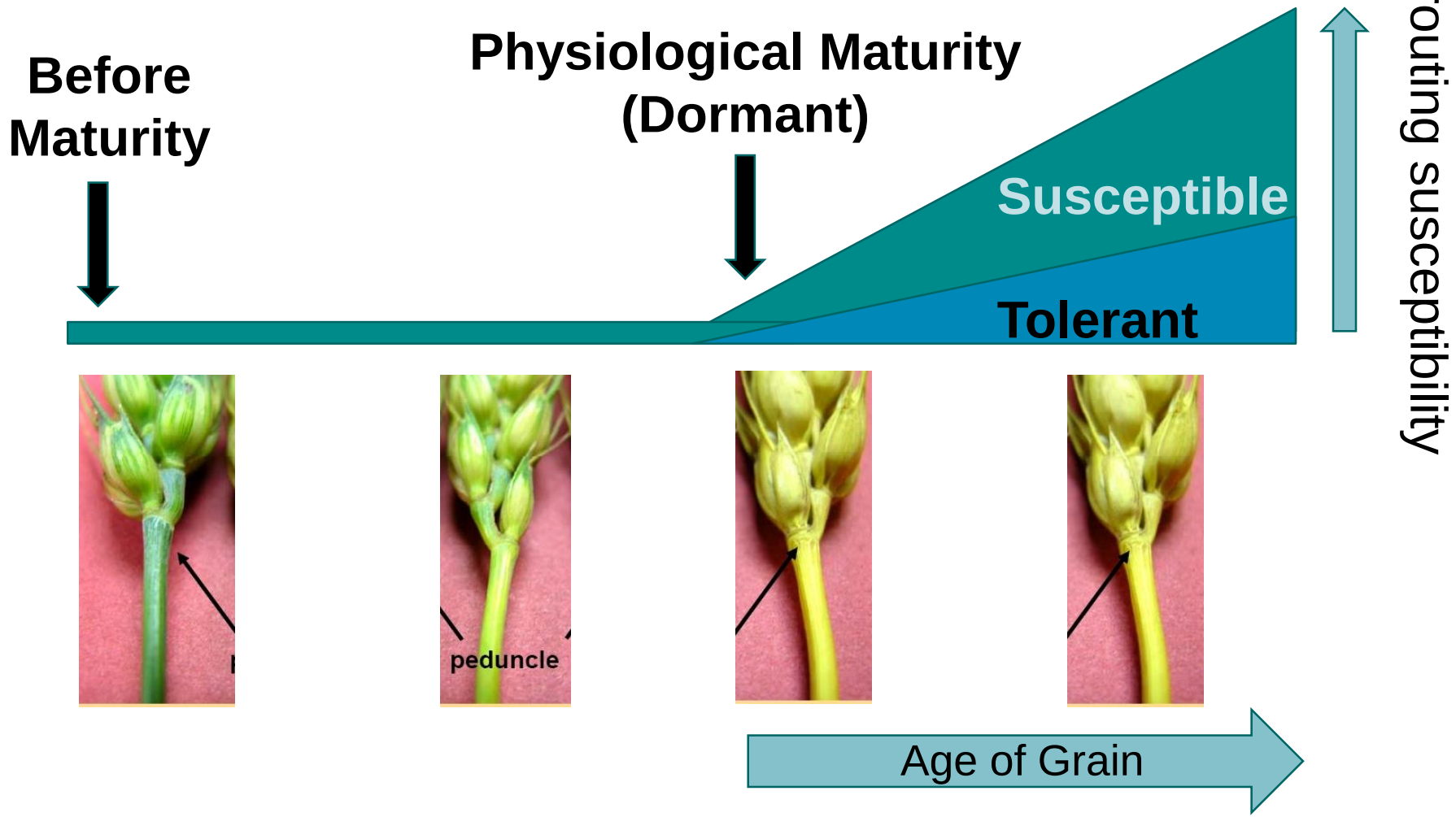
Wheat Seed Dormancy

The inability to germinate even under **favorable environmental conditions**

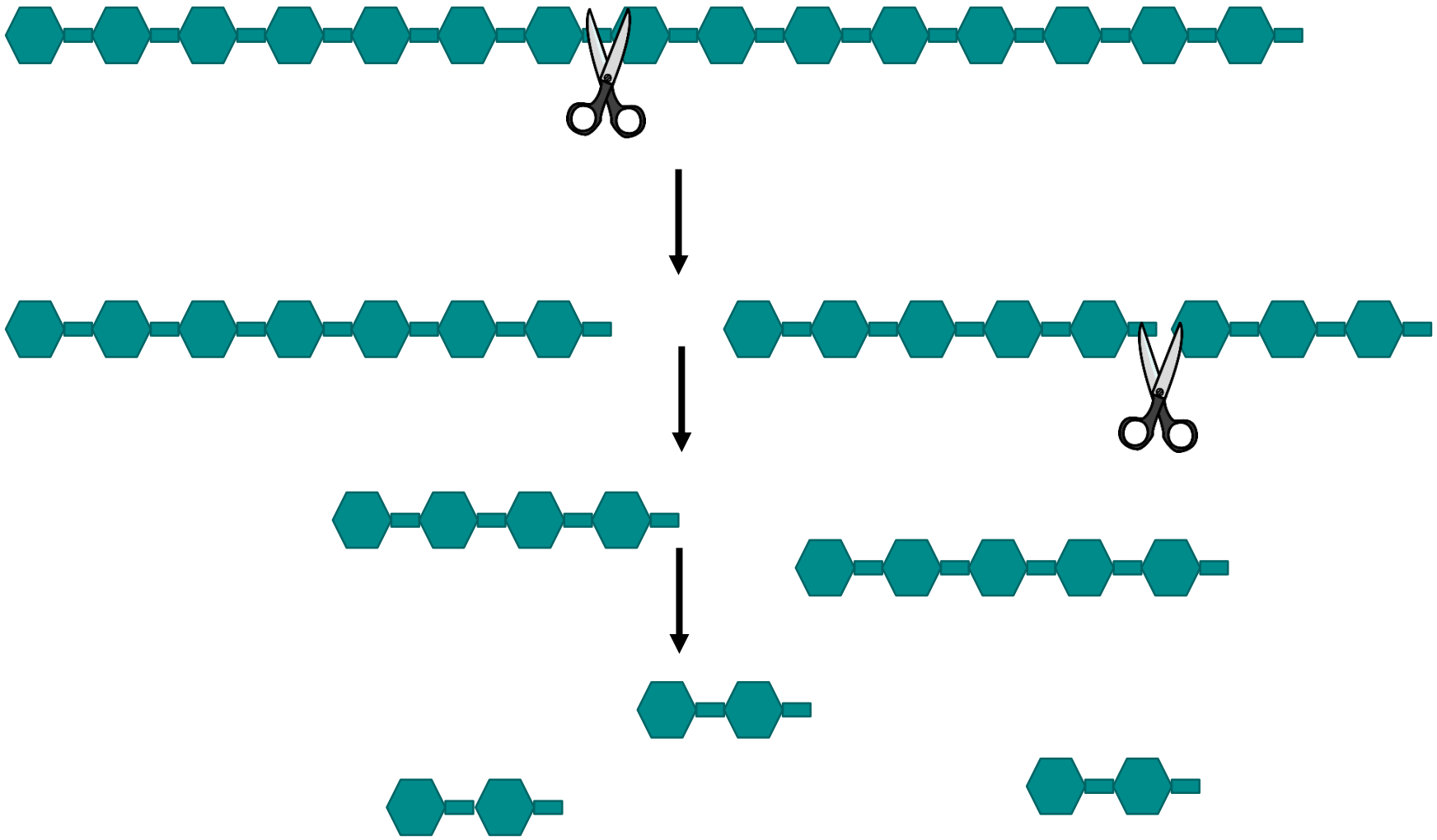


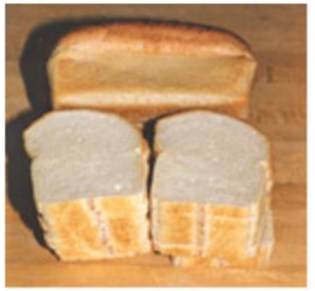
After-ripening
Cold Imbibition

Susceptibility to preharvest sprouting depends on maturity date



PHS is a result of a-amylase activity breaking down starch chains

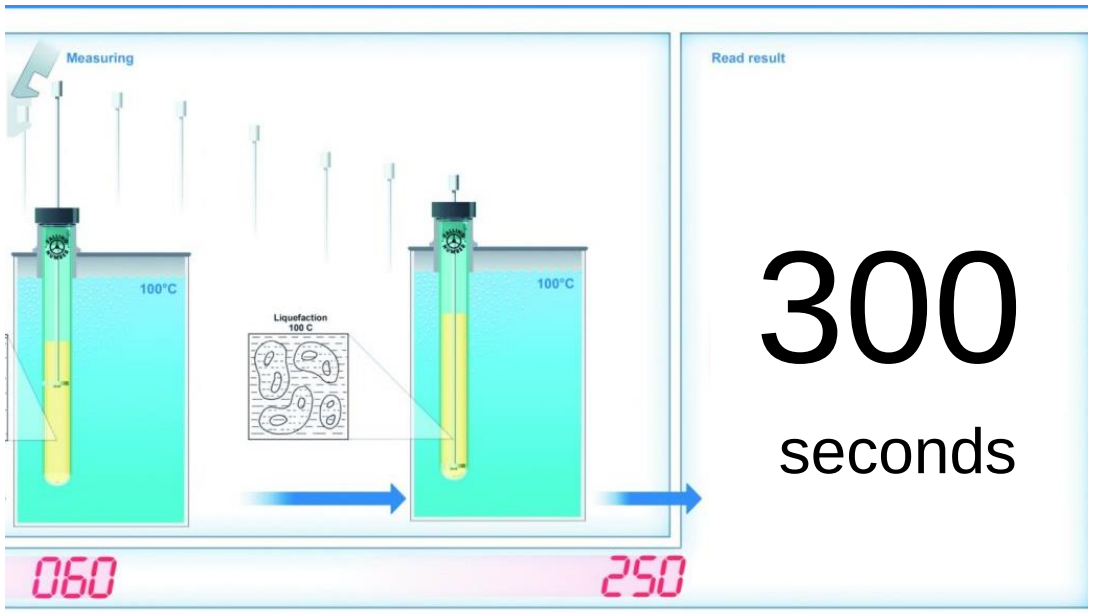




Sound

Severely Sprouted

Hagberg-Perten Falling Number: The Industry Standard to Measure PHS



www.perten.com

Farms.com

Low FN is associated with low end use quality

You are Not Alone - 2018 Preharvest Sprouting

England

Tobias Barber @ekte_Toby — Following

I guess this is what you'd call pre-harvest sprouting #Harvest18

2:21 PM - 25 Aug 2018

Jim Thompson @jimt_farmer — Follow

Think rain has stopped play #wheatharvest18 @AllpressF @LumleySean @coostiebarrey @chrisbettinson2

9:53 AM - 27 Jul 2018

1 Retweet 8 Likes

Kansas

Kyler Millershaski @Shaski92 — Follow

I'm always happy to have rain, but not the view I want during #WheatHarvest18 #kswx

3:36 PM - 22 Jun 2018

5 Retweets 37 Likes

Chris Cu11an @ChrisCullan1 — Follow

On the edge. #wheatharvest18

5:06 PM - 12 Jul 2018

New York

Shantel A. Martinez @s_amealia

A lot of hopes and dreams were crushed this week. Mon. I finished my harvest for PHS trials, then it rained for 2.5 days. Today we all went out to harvest the barley trials, SPROUTED. Went to harvest my mapping pop. for planting seed, SPROUTED. Welcome to NY summers?

5:37 PM - 26 Jul 2018

1 Retweet 6 Likes

2 1 6

Nebraska

North Dakota (2017)

Canada

Fermes Chauvin Farms Ltd. @MoeChauvin — Follow

#wheatharvest18 started here at fermeschauvinfarms.com in StoneyPoint. Decent yields for no rain ... #OntAg #AgMoreThanEver #goodineverygrain

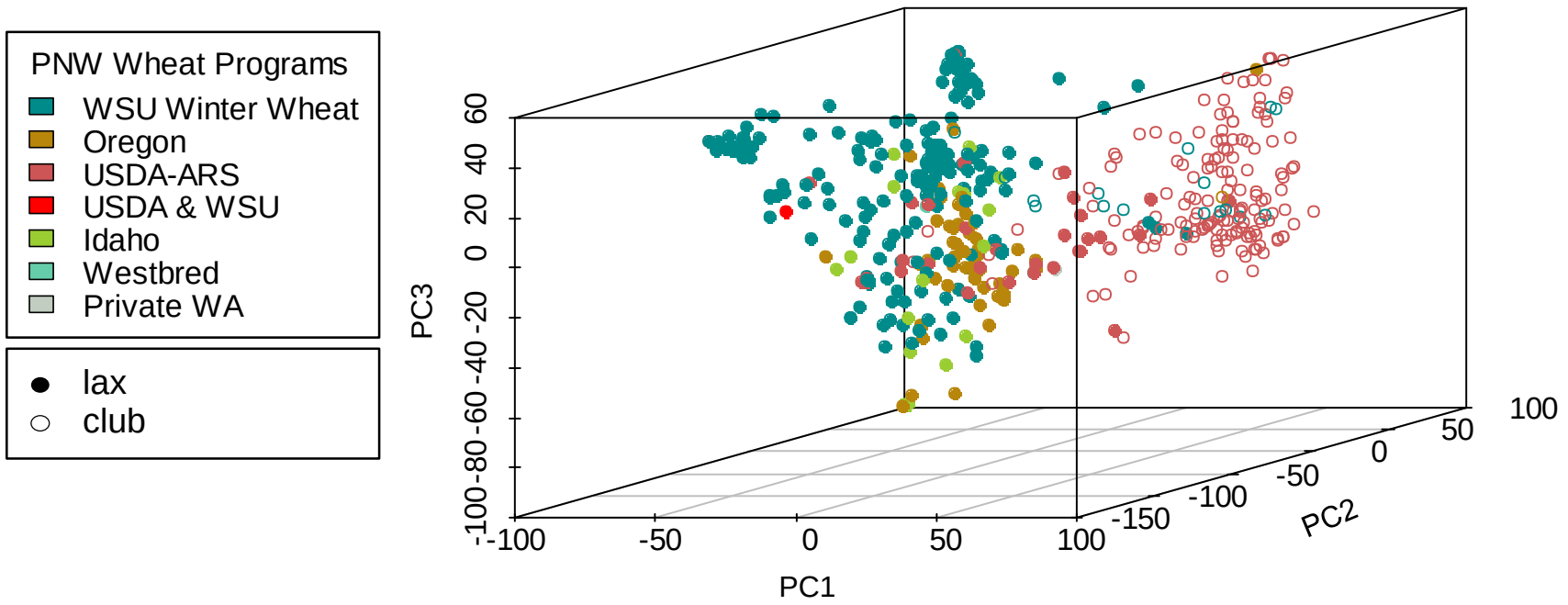
4:51 PM - 5 Jul 2018

3 Retweets 37 Likes

Mory Rugg @NPWheat — Following

This is why I screen for Phs

Improving Preharvest Sprouting in the PNW: Understanding the Genetic Tolerance that Exists in the Current Breeding Programs



PHS

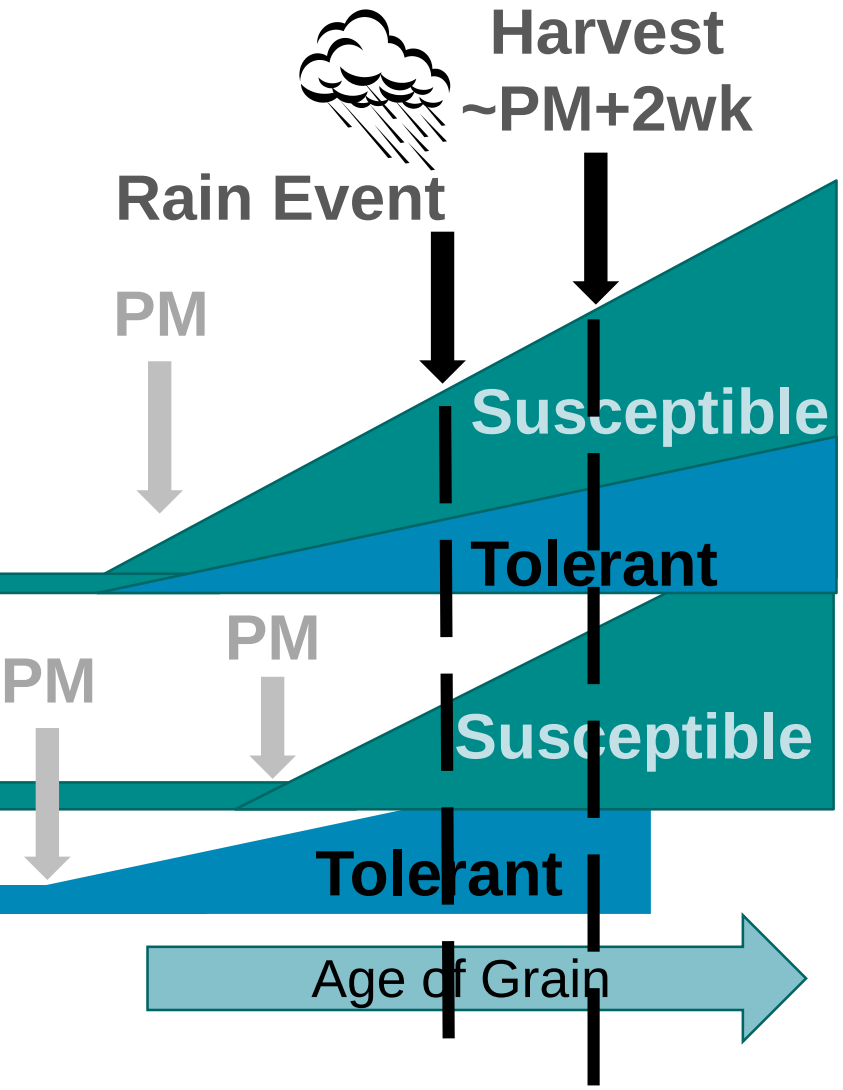
Falling Number

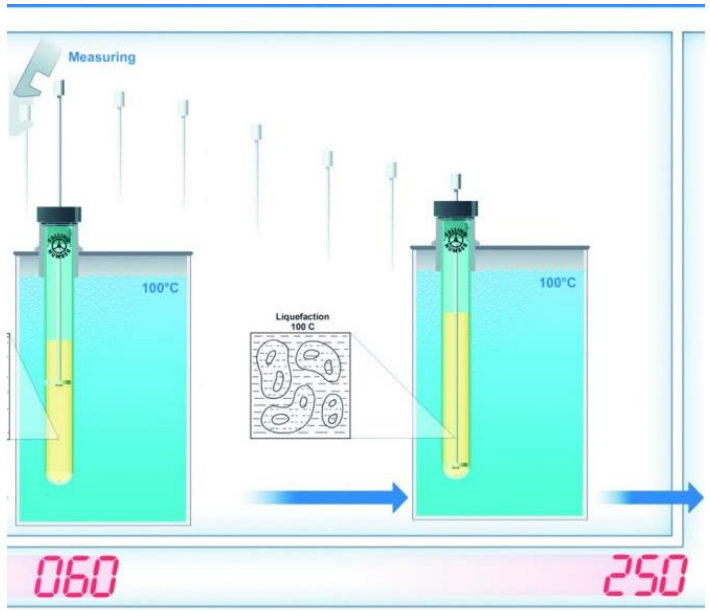
Spike Wetting Test

The panel is derived from at least six white winter wheat breeding programs.

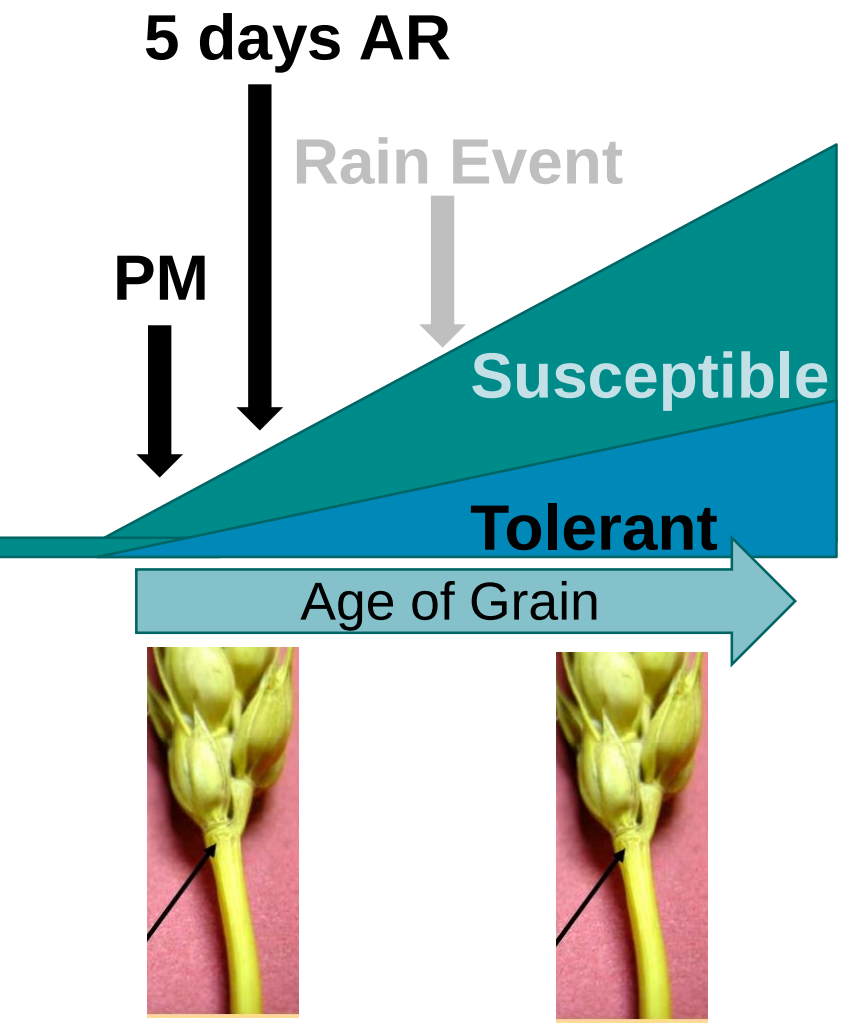
Jernigan and Godoy et al., 2018 | Martinez et al., 2016

Falling Numbers Test:
Samples are Harvested at “Harvest Maturity”





Greenhouse Spike Wetting Test: Samples are Harvested at Physiological Maturity

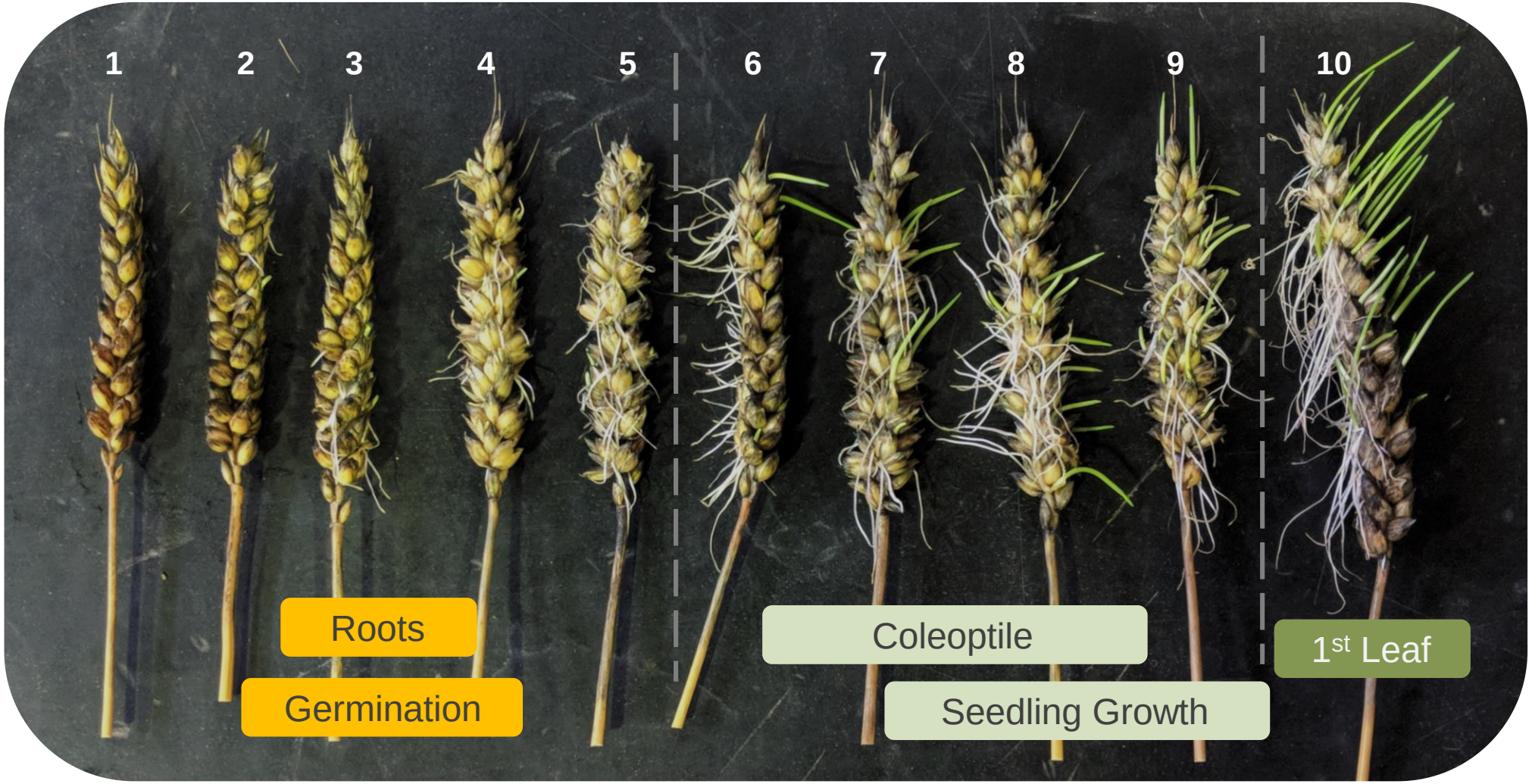


Misted 6 sec / min

Scored every 24 hrs for 7 days

Visible Sprout Scored

PHS Tolerant ⟷ PHS Susceptible

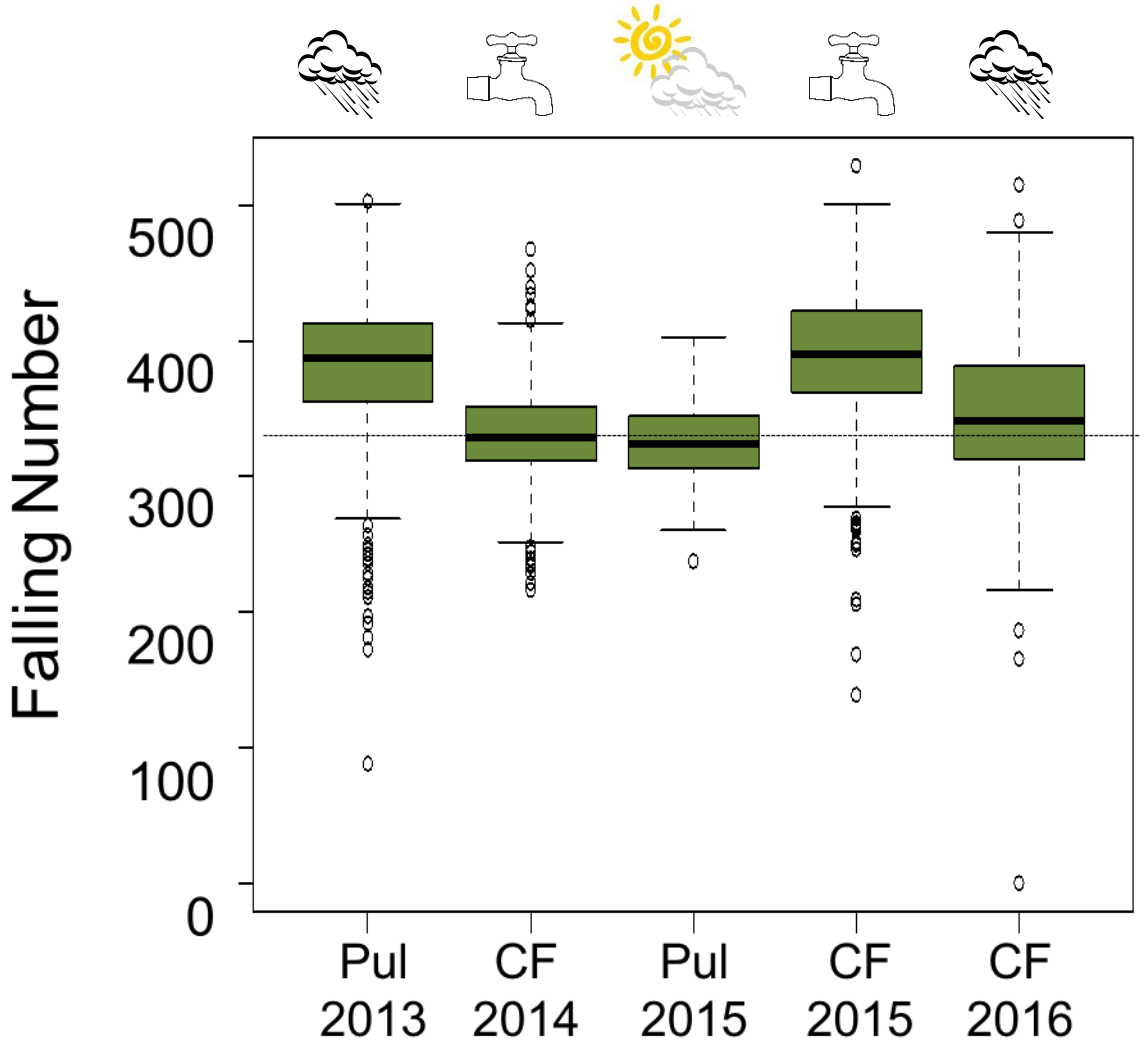


McMaster & Derera *et al*., 1976

Hypothesis

If sprouting is the main cause of low FN, then similar loci should be detected based on Falling Numbers and on the appearance on visible sprouting in spike wetting tests through association mapping.

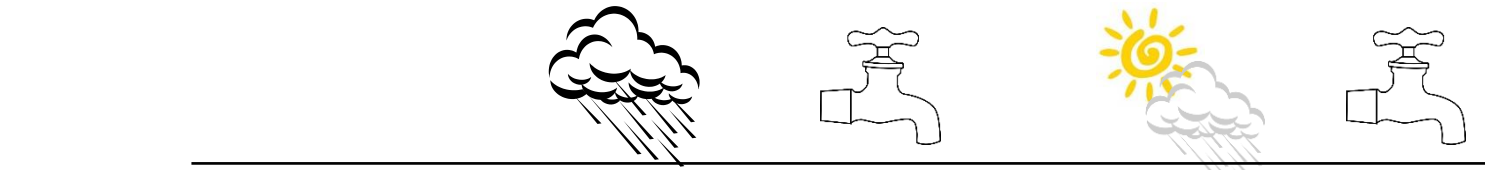
The FN Trait Was Tested Over 5 Environments, 3 Different Events



Pul: Pullman

CF: Central Ferry

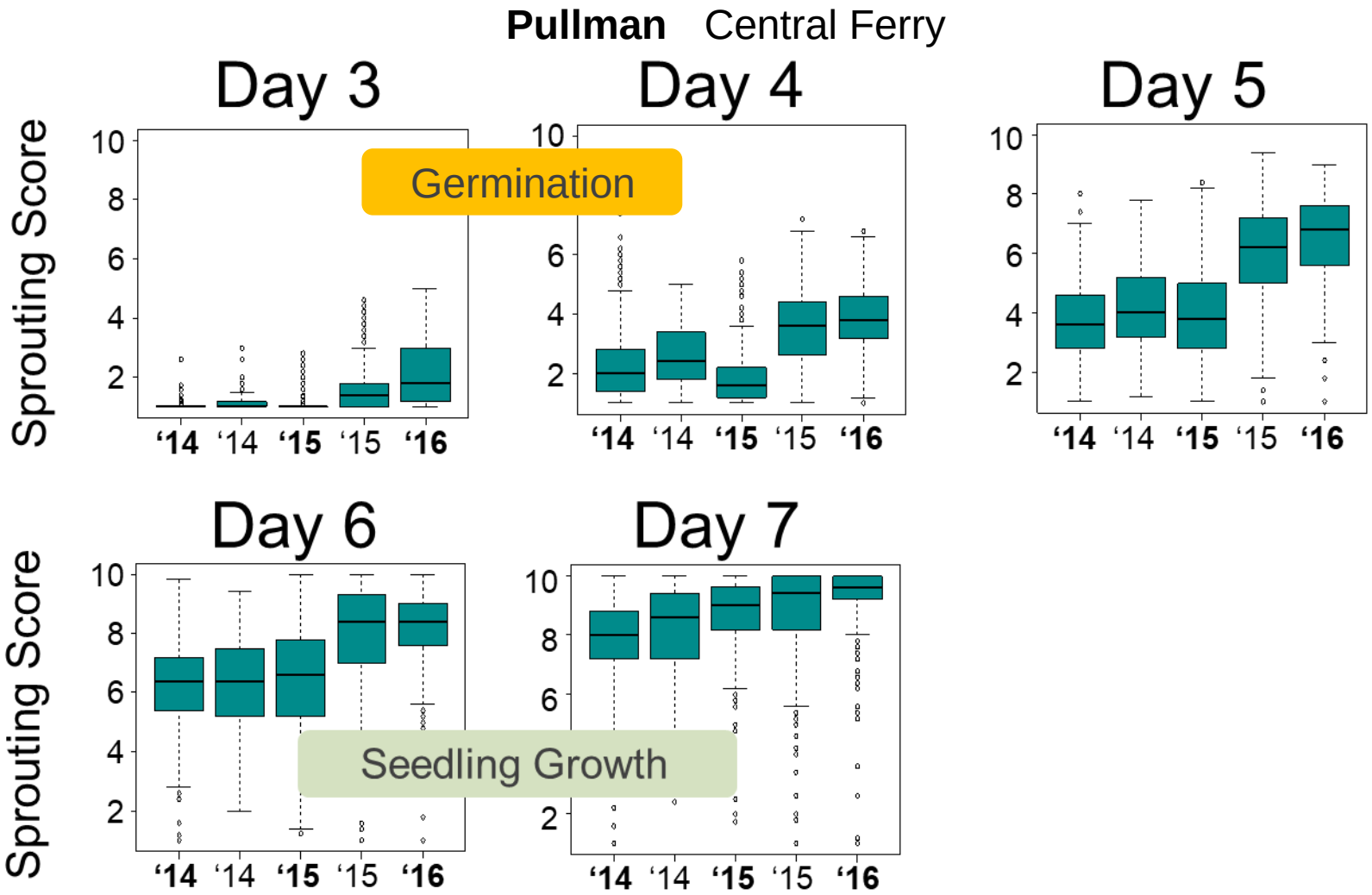
The environment has a large affect on the FN response



	Pul13	CF14	Pul15	CF15
CF14	0.29**			
Pul15	0.23**	**0.42****		
CF15	0.23**	0.29**	0.29**	
CF16	0.33**	**0.46****	0.30**	0.34**

**: $p \leq 0.001$ * : $p < 0.05$

Visible Sprouting Was Tested Over 5 Environments, Same GH Rain Event



Visible Sprout Correlations Across Environments Were as Good as Other SWT Studies

Day 6 Seedling Growth

	CF14	Pul16	Pul14	CF15
Pul16	0.39**			
Pul14	0.39**	0.29**		
CF15	0.38**	0.30**	**0.40****	
Pul15	0.34**	0.16*	**0.46****	0.36**

**: $p \leq 0.001$ * : $p \leq 0.05$

Kulwal et al., 2012; Jaiswal et al., 2012; Ogbonnaya et al., 2008; Zhou et al., 2017

The Correlations Between FN and Visible Sprout are Not Highly Negative

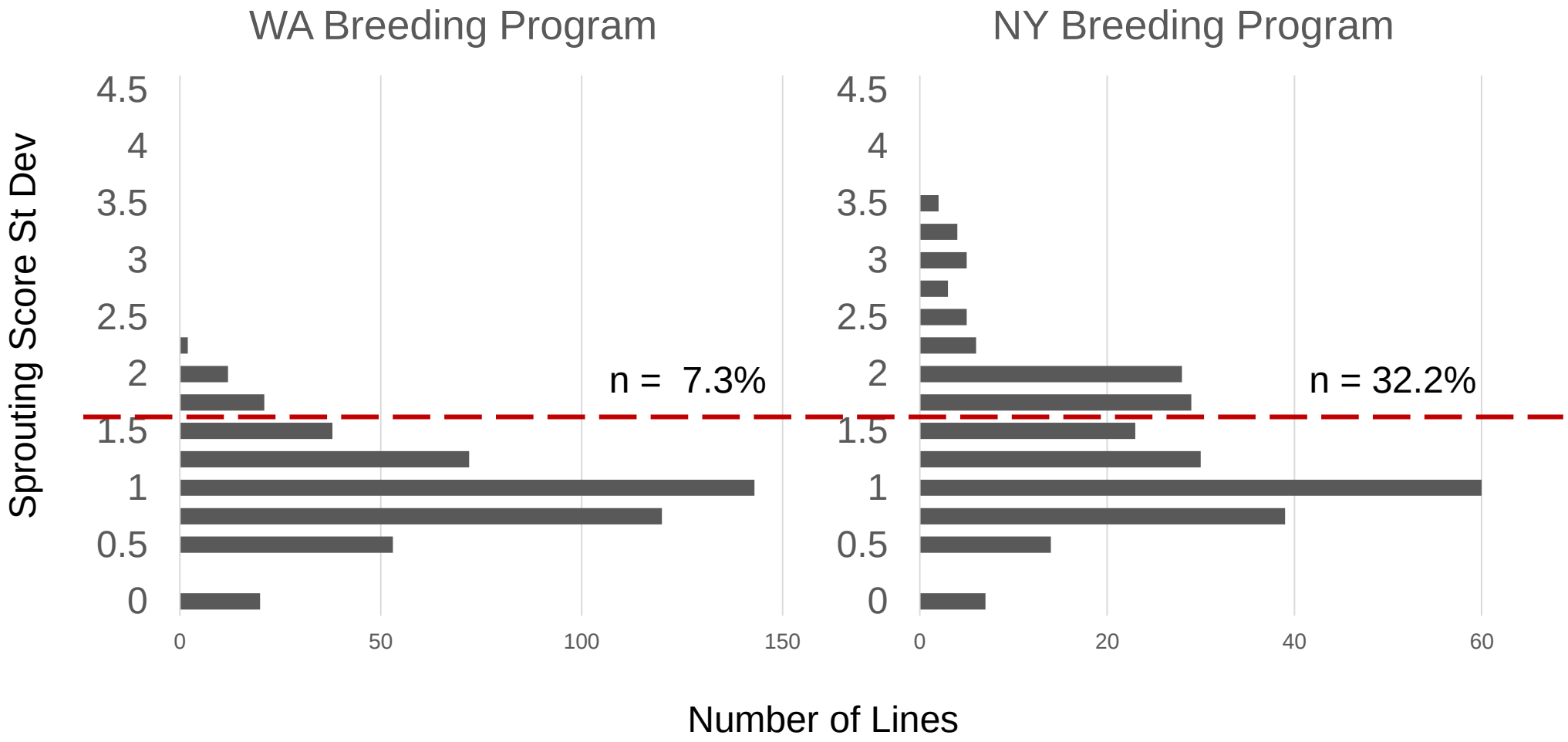
	Germination			Seedling Growth		
	3 days	**4 days**	**5 days**	**6 days**	**7 days**	**SI**
 **Pul13**	-0.16**	-0.24**	-0.17**	-0.18**	-0.20**	-0.21**
 **CF14**	-0.07	-0.09*	-0.06	-0.09	-0.10*	-0.10*
 **Pul15**	-0.07	-0.13*	-0.12*	-0.12*	-0.17**	-0.15**
 **CF15**	-0.09	-0.04	0.00	0.01	0.00	-0.01
 **CF16**	-0.17**	-0.19**	-0.18**	-0.17**	-0.17**	-0.19**

**: $p \leq 0.001$ * : $p \leq 0.05$

-0.80** -0.83**

Rasul et al., 2009; Jiménez et al., 2016

When Implementing Spike-Wetting Tests in a Breeding Program: The Germplasm / Environment Could Affect the Variance



Cornell University & Washington State University, unpublished

Genome-wide Association Study of FN and Visible Sprout

15,229 polymorphic markers | 21 chromosomes | 469 accessions

9 *QFN.wsu*

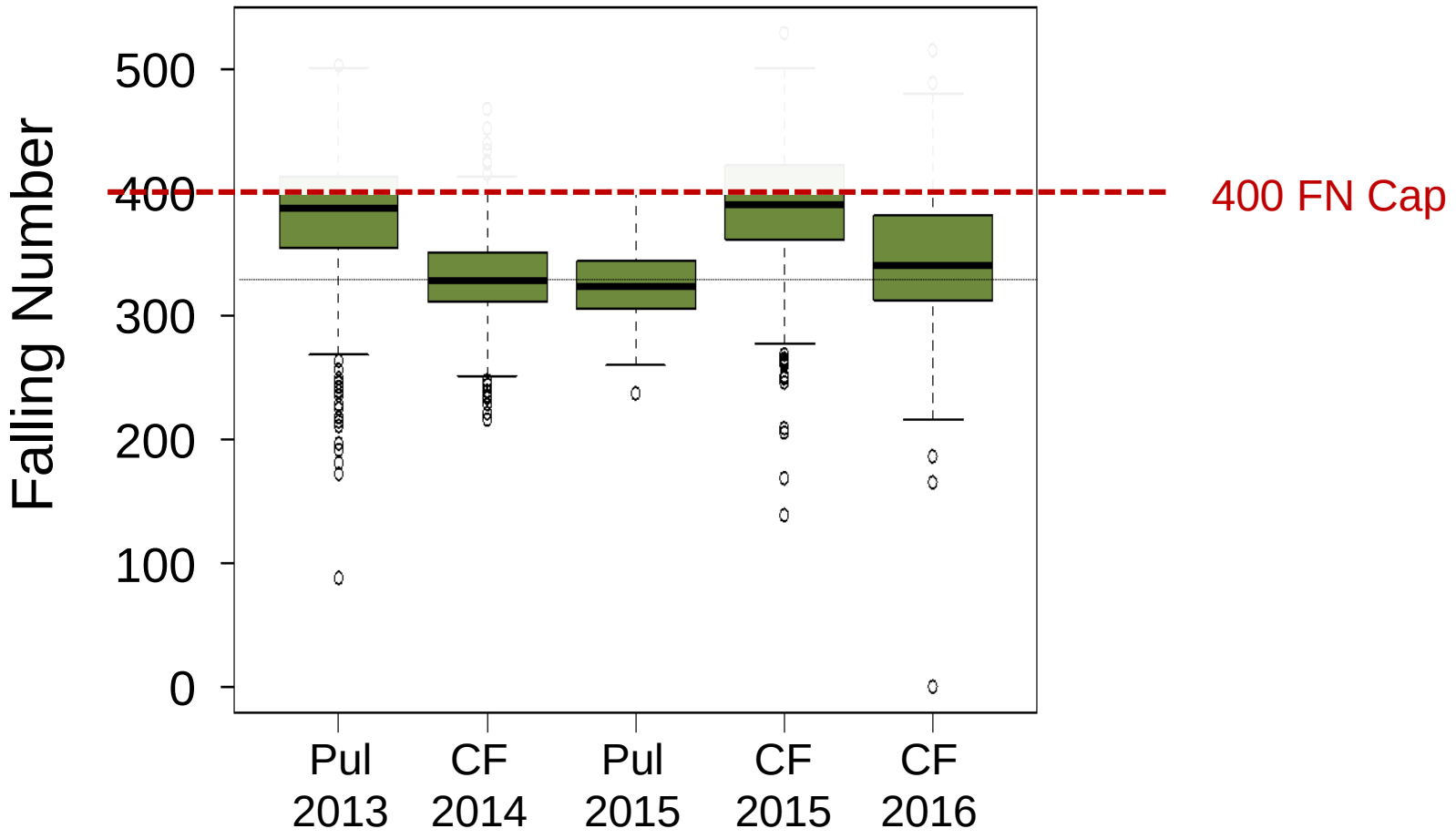
CF16	Pul15	CF15
3	3	0
Pul13		**CF14**
3		0

34 *QPHS.wsu*

Germination | Seedling Growth

Day 3 Day 4	Day 5	Day 6 Day 7	SI
12	6	13	3

Will Only Looking at the Low FN Values Give Different QTN?



Genome-wide Association Study of FN and Visible Sprout

15,229 polymorphic markers | 21 chromosomes | 469 accessions

2 QFN.wsu 9 *QFN.wsu*

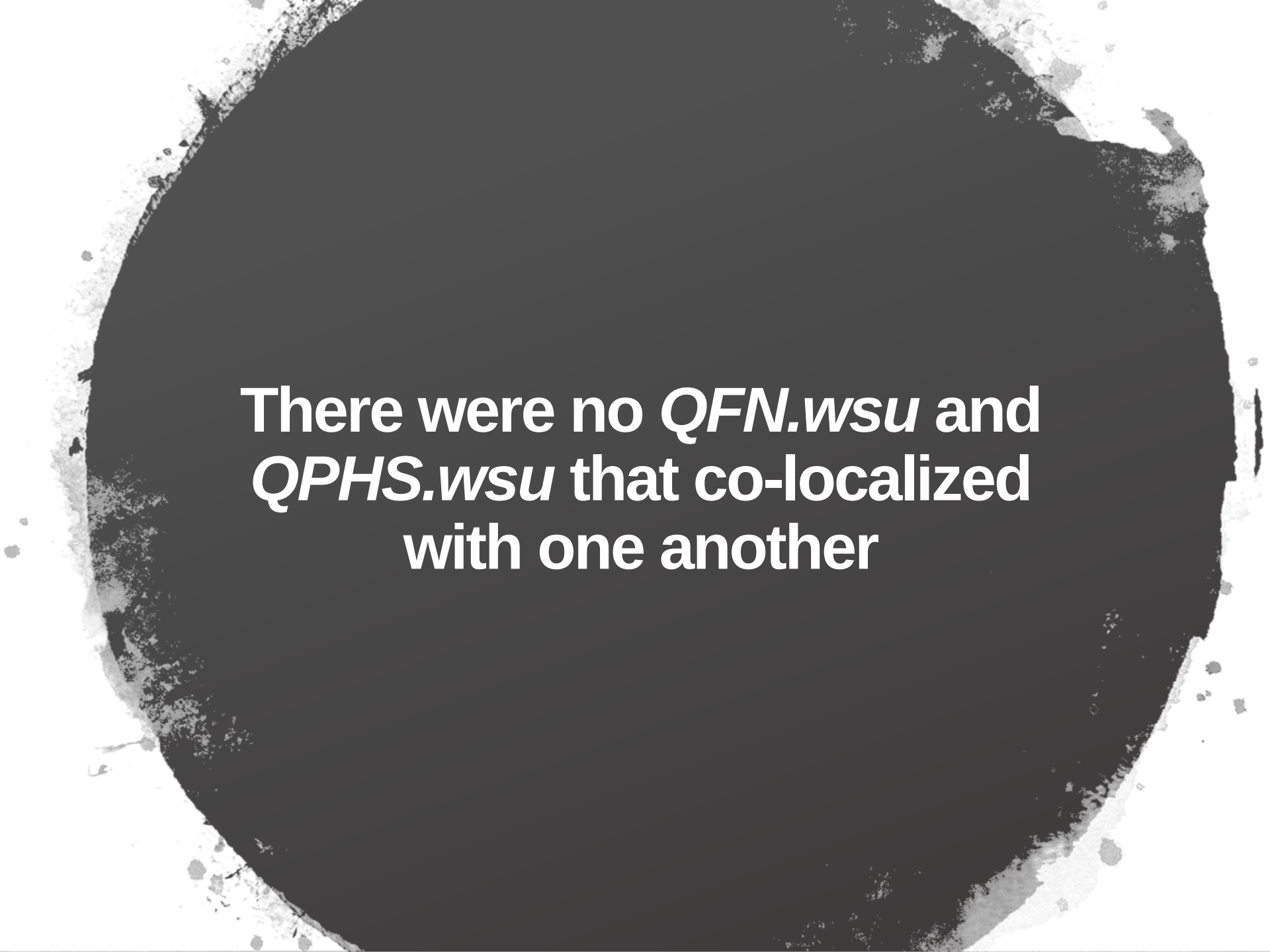


CF16	Pul15	CF15
3+2	3	0
Pul13		**CF14**
3		0

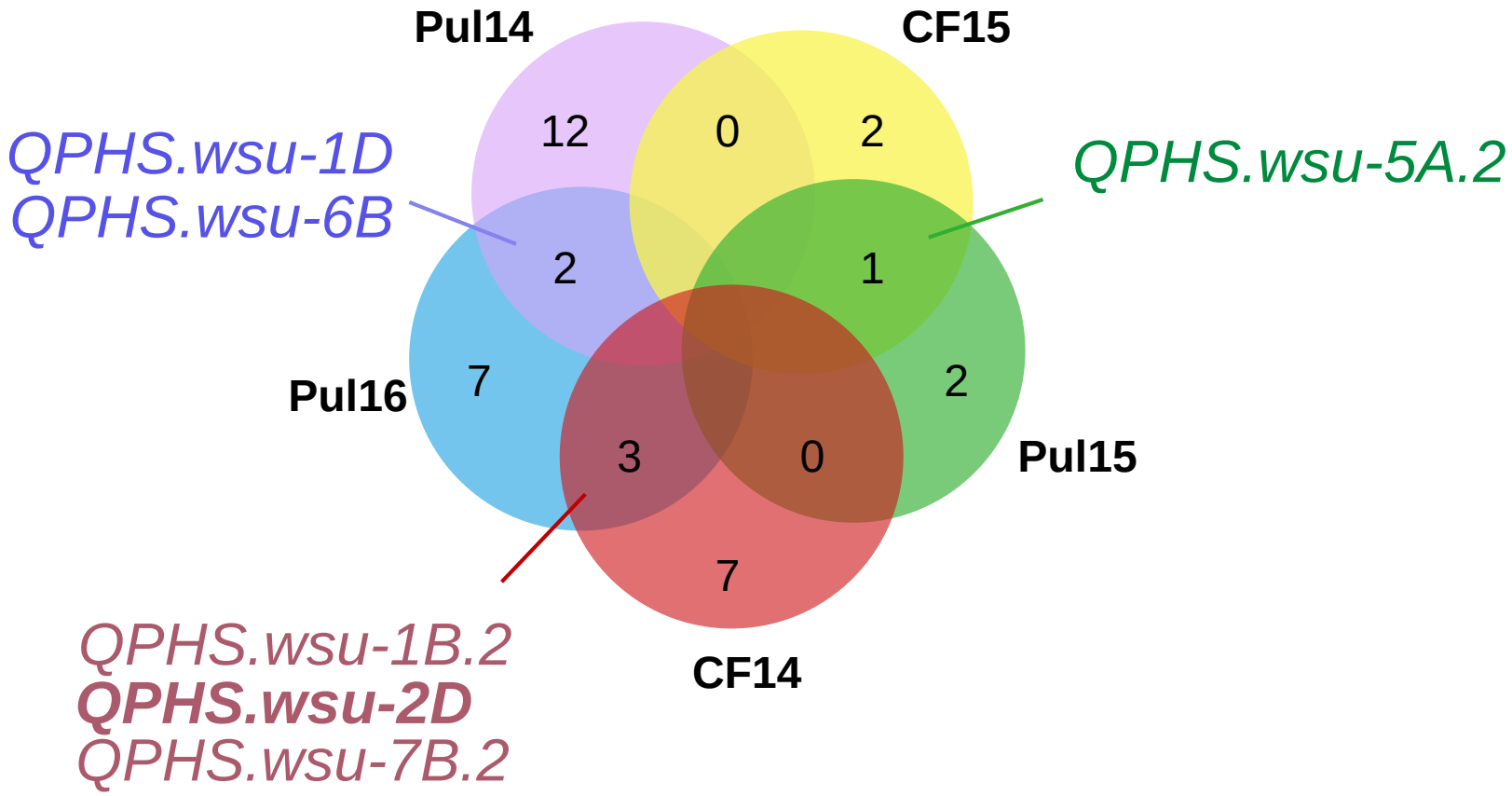
34 *QPHS.wsu*

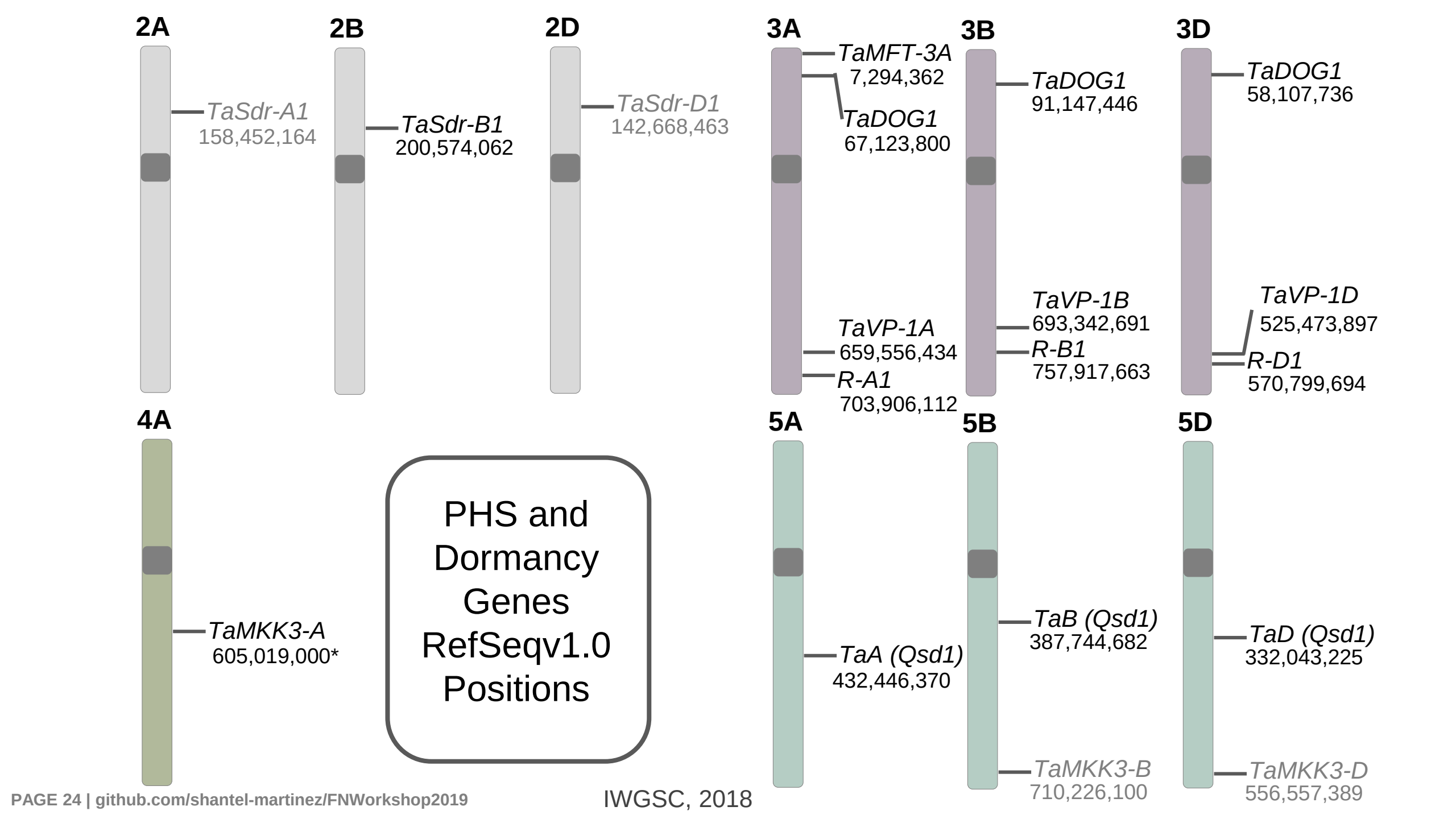
Germination | Seedling Growth

Day 3 Day 4	Day 5	Day 6 Day 7	SI
12	6	13	3


There were no *QFN.wsu* and *QPHS.wsu* that co-localized with one another

Visible Sprouting QTL *QPHS.wsu* Across Environments

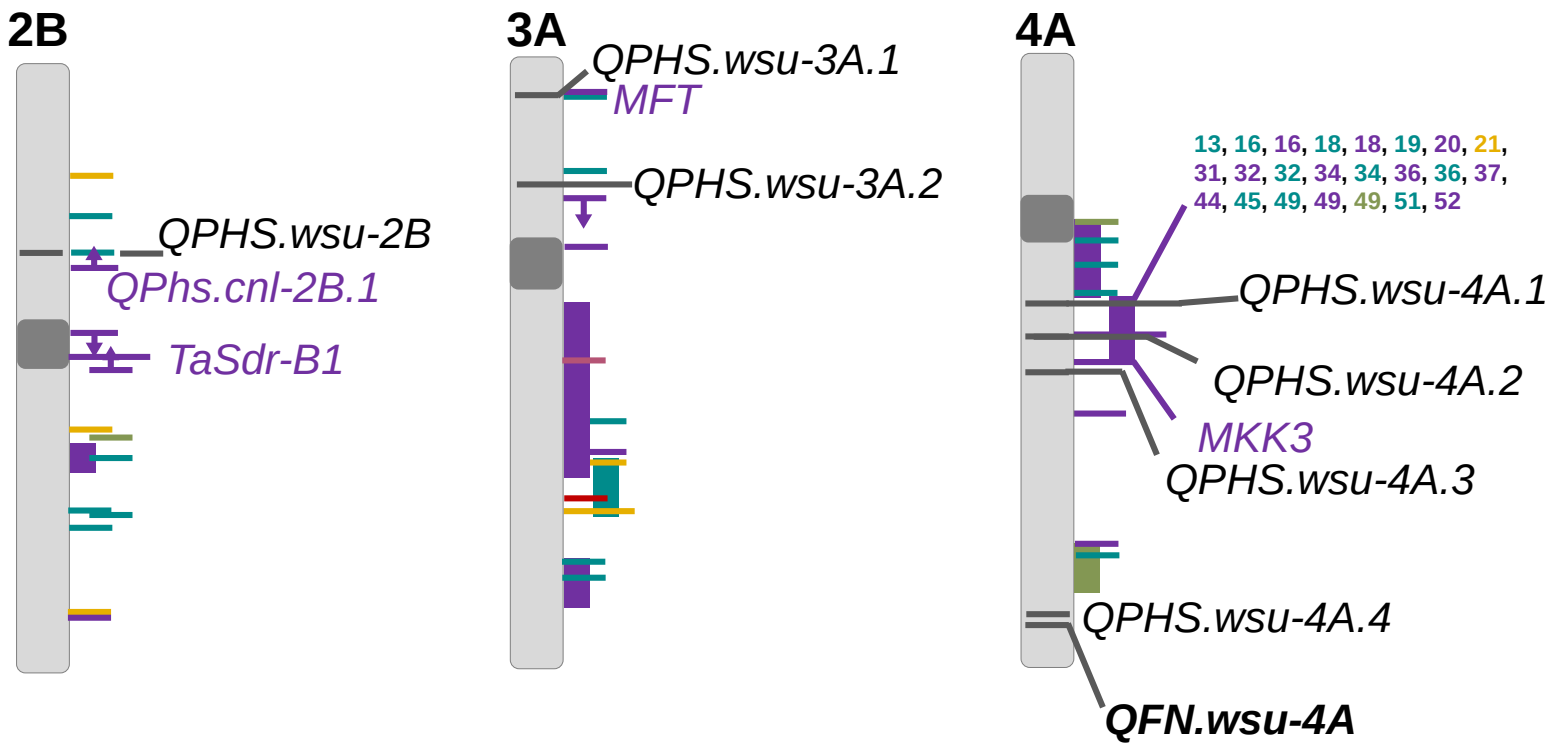



2A
TaSdr-A1
158,452,164
2B
TaSdr-B1
200,574,062
2D
TaSdr-D1
142,668,463
3A
TaMFT-3A
7,294,362
TaDOG1
67,123,800
TaVP-1A
659,556,434
R-A1
703,906,112
3B
TaDOG1
91,147,446
TaVP-1B
693,342,691
R-B1
757,917,663
3D
TaDOG1
58,107,736
TaVP-1D
525,473,897
R-D1
570,799,694
4A
TaMKK3-A
605,019,000*
PHS and Dormancy Genes RefSeqv1.0 Positions
5A
TaA (Qsd1)
432,446,370
5B
TaB (Qsd1)
387,744,682
TaMKK3-B
710,226,100
5D
TaD (Qsd1)
332,043,225
TaMKK3-D
556,557,389

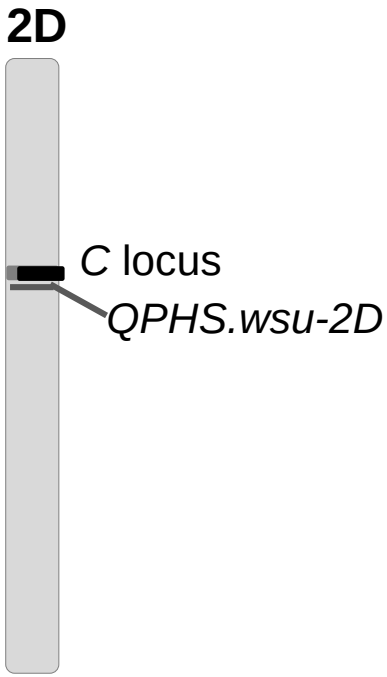
IWGSC, 2018

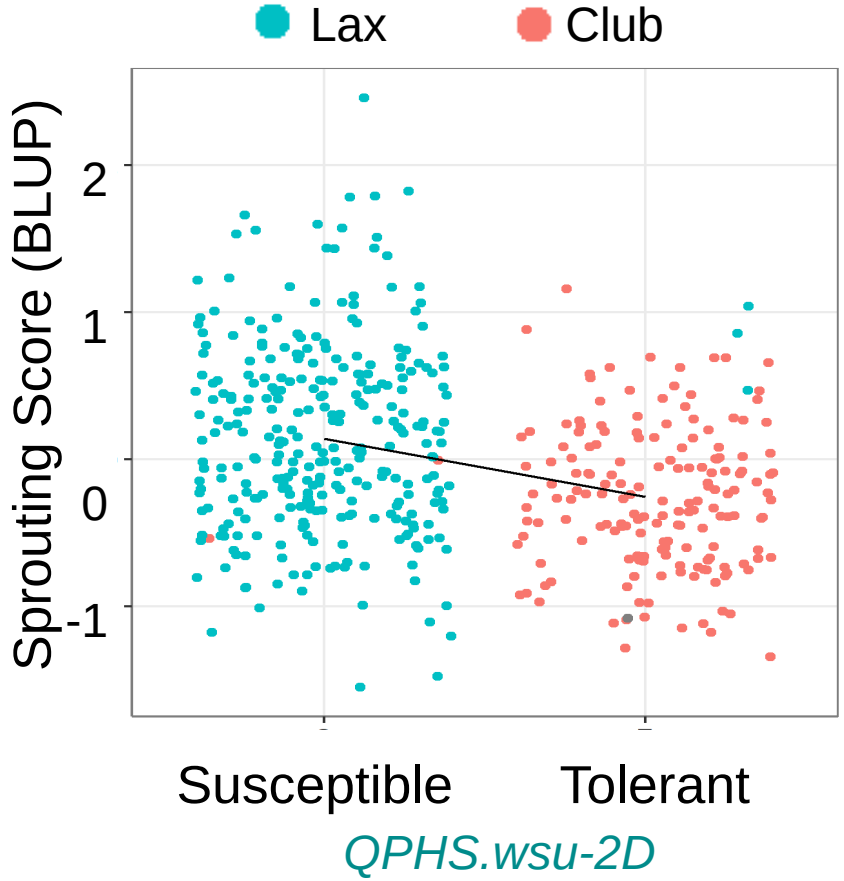
Visible Sprouting QTL Located Near Known PHS Genes

Sprouting Assay Falling Numbers Grain Color Martinez et al. QTN
Dormancy Assay Quality LMA



Strongest PHS QTL, *QPHS.wsu-2D*, Close to the *Compactum* (*C*) Locus





Photos by T. DeMacon | Johnson et al., 2008

2 of the 11 *QFN.wsu* appear to be unique

10 of the 34 *QPHS.wsu* appear to be unique

The others were found near other known PHS-related loci

Breeding for PHS Conclusion

GWAS for FN and visible sprout detected different QTN, although both co-localized with known PHS-related loci

FN is a measure of a-amylase activity. The lack of correlation between FN and visible sprout may mean that a-amylase is regulated differently with respect to the timing of germination in different varieties.

The club *C* locus was linked to the strongest *QPHS.wsu-2D* QTL

github.com/shantel-martinez/FNWorkshop2019

Panel Topic:

How do we make these markers/QTL useful in breeding programs

How do we turn this into a tool the breeders can use now