Genomic Selection to Increase the Rate of Genetic Gain of Intermediate Wheatgrass (*Thinopyrum intermedium*)

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Introduction

Intermediate Wheatgrass (IWG) is a promising perennial species that is being domesticated as a grain and biomass crop. Initial breeding work began at the Rodale Institute, Kutztown, PA, in the 1980's. In 2002, The Land Institute, Salina, KS, began breeding IWG using selections made by the Rodale Institute [1]. Early and contemporary work has been encouraging, with seed yield increasing 77% over two cycles of selection, and seed mass increasing 23% [1]. While these gains have been important, if the current rate of gains holds, it is estimated that it would take between 24 and 110 years of breeding to reach the yield and seed size of annual wheat [1]. There are numerous challenges to breeding IWG including:

- Large genome size 12.6 Gb
- Allohexaploid (2n = 6x = 42)
- Outcrossing and heterozygous

As a perennial, breeding IWG is a time intensive process, with a typical breeding cycle length of two years (Figure 1).
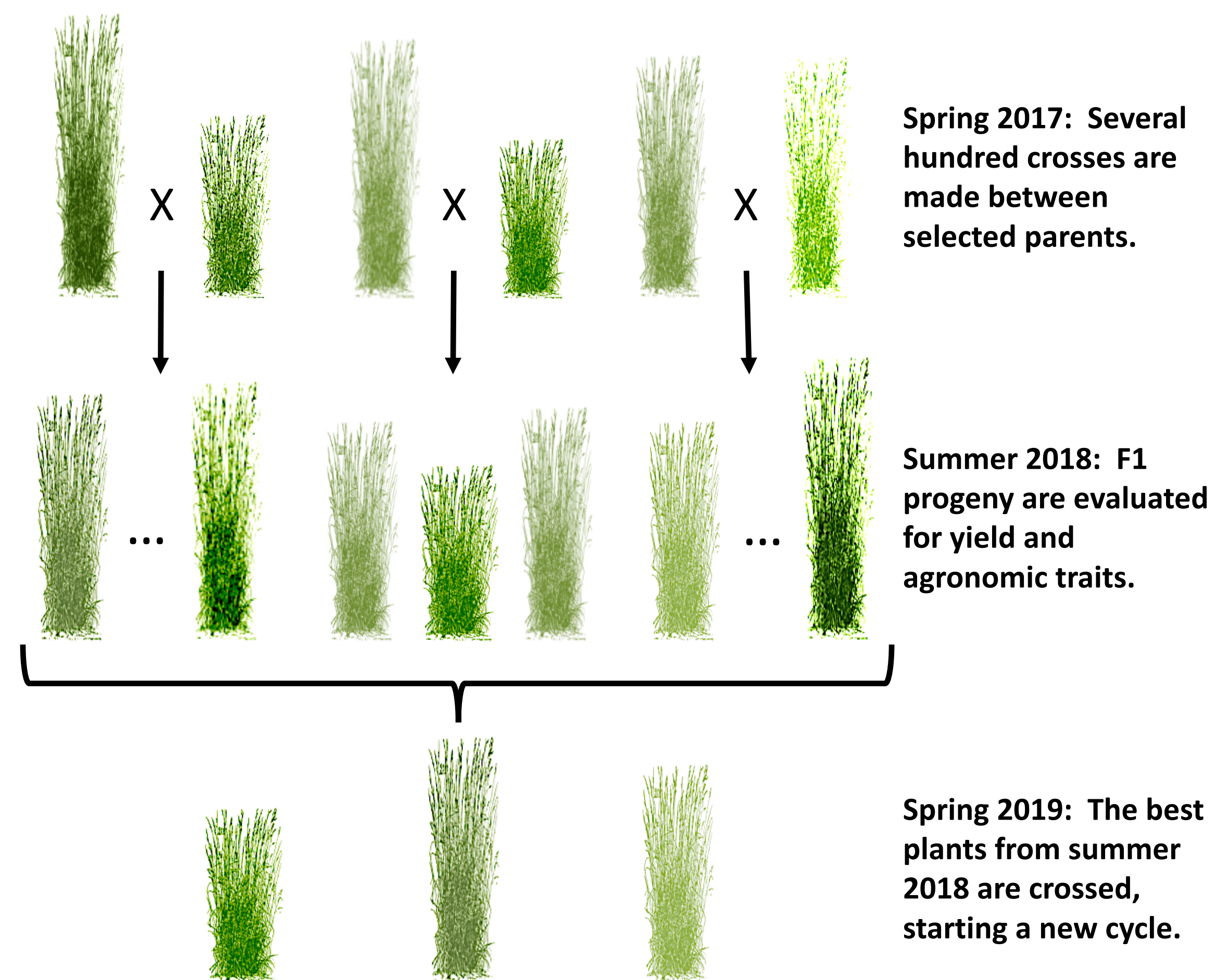

Spring 2017: Several hundred crosses are made between selected parents.

Summer 2018: F1 progeny are evaluated for yield and agronomic traits.

Spring 2019: The best plants from summer 2018 are crossed, starting a new cycle.

Figure 1. Two year phenotypic breeding cycle for Intermediate Wheatgrass.

Recently, genomic selection (GS) has been proposed as a way to increase the rate of genetic gain in IWG. Work by Zhang et al. [2] showed high predictive ability of GS for several agronomic traits, including seed weight and biomass. In addition to high predictive ability, GS could be used to drastically reduce the length of the breeding cycle, allowing for intermating of selected progeny each year (Figure 2) and resulting in a breeding cycle that is twice as fast. Given the potential of GS to increase the rate of genetic gain, we examined how GS could be applied and developed methods to apply GS to The Land Institute IWG breeding program.
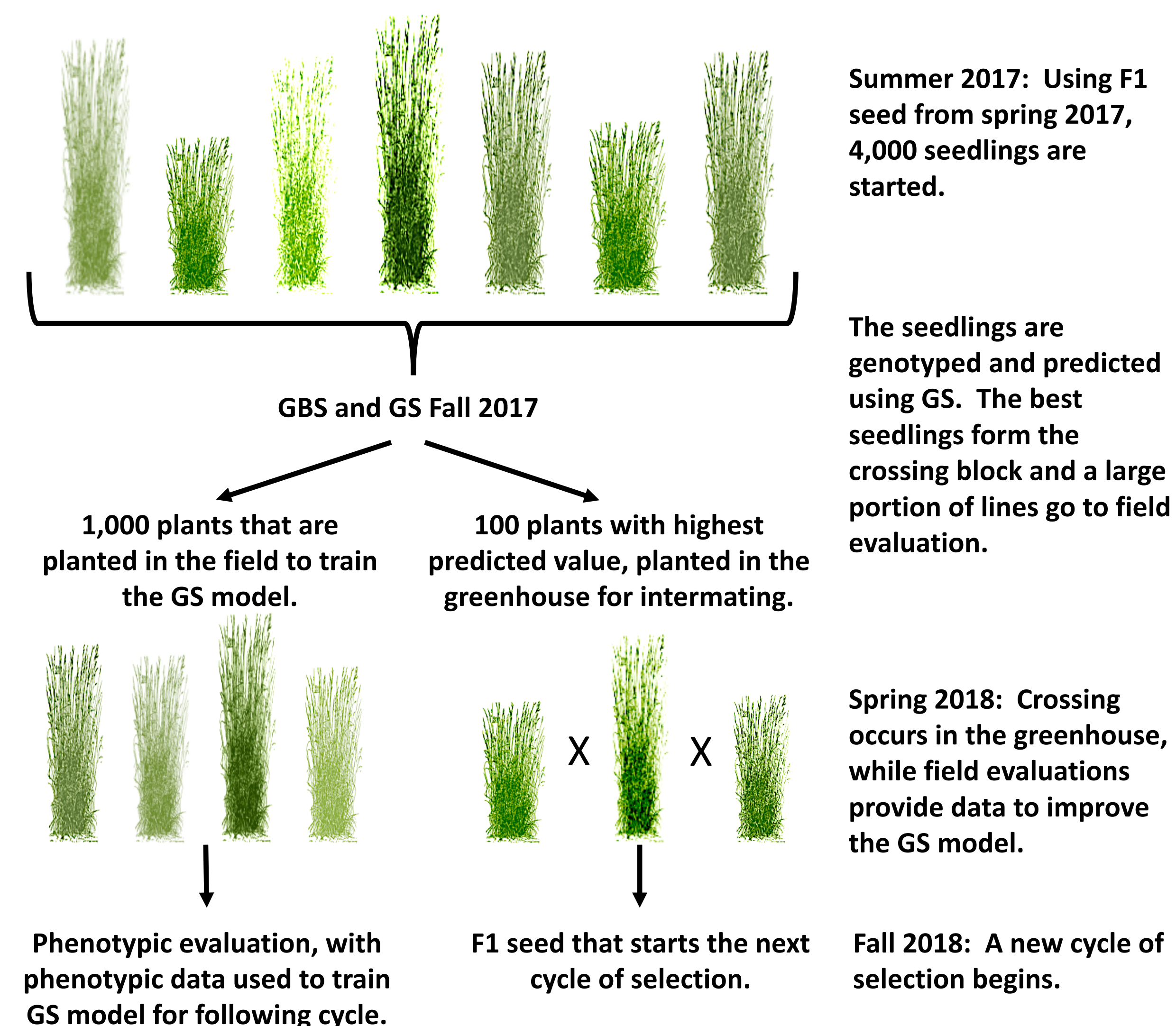

Summer 2017: Using F1 seed from spring 2017, 4,000 seedlings are started.

The seedlings are genotyped and predicted using GS. The best seedlings form the crossing block and a large portion of lines go to field evaluation.

GBS and GS Fall 2017

1,000 plants that are planted in the field to train the GS model.

100 plants with highest predicted value, planted in the greenhouse for intermating.

Spring 2018: Crossing occurs in the greenhouse, while field evaluations provide data to improve the GS model.

Phenotypic evaluation, with phenotypic data used to train GS model for following cycle.

F1 seed that starts the next cycle of selection.

Fall 2018: A new cycle of selection begins.

Figure 2. Application of genomic selection to Intermediate Wheatgrass breeding. The genomic selection model predicts plant performance, with the best plants forming the parents for the next cycle. By simultaneously evaluating material in the field and crossing plants, the time to complete the cycle can effectively be cut in half.

Materials and Methods

Cycle 6 of The Land Institute IWG breeding program formed the training population, genotyping-by-sequencing (GBS) was used for marker discovery and genotyping. The population included:

- 3,658 genets in a single replicate row-column design.
- Genets were phenotyped for 46 traits over two years.
  Seed yield, spike fertility, seed mass, …, stem strength.
  For each trait, 1,880-2,400 observations were recorded.
- 2,974 plants genotyped using GBS.

Using the available data, best linear unbiased predictors (BLUPs) were calculated for each trait. Heritability (h^2) was calculated using the genomic marker matrix. Cross-validation proceeded by leaving entire parents out of the training set, to prevent bias of close relatives predicting performance, and several GS models were tested to identify the best performing models.

Table 1. Heritability and cross-validation prediction accuracies for phenotypic traits from Cycle 6 training population. Cross-validation accuracy is reported as the correlation coefficient between the phenotypic best linear unbiased predictor and the predicted value.

Trait	h^2	r^2
Free Threshing 2016	0.61	0.45[†]
Free Threshing 2017	0.50	0.91[†]
Spike yield in grams 2017	0.36	0.89
Milligrams per seed 2016	0.58	0.86
Plant height 2016	0.63	0.80

[†]Further work is investigating the discrepancy between 2016 and 2017 model predictions.

Results and Discussion

Heritability across all traits ranged from 0.28-0.67, and the cross-validation prediction accuracy was high for all traits ranging from 0.45-0.93 (Table 1). Across multiple GS models, the prediction values among genets were quite stable, with the Gaussian Kernel and rrBLUP having the most variation in prediction. Even with this variation, for any individual trait the minimum correlation between these two models was $r^2 > 0.94$ (Figure 3).
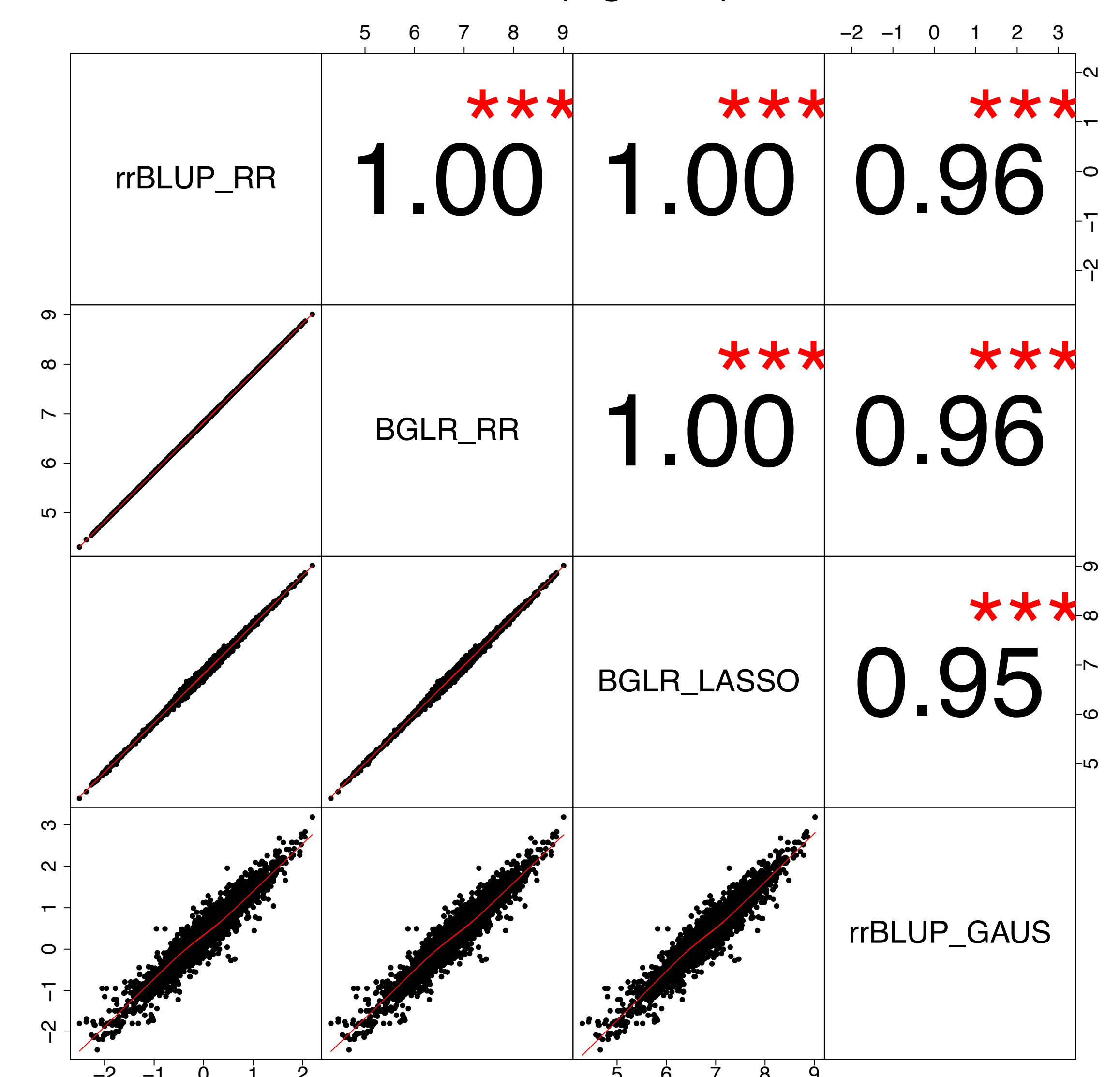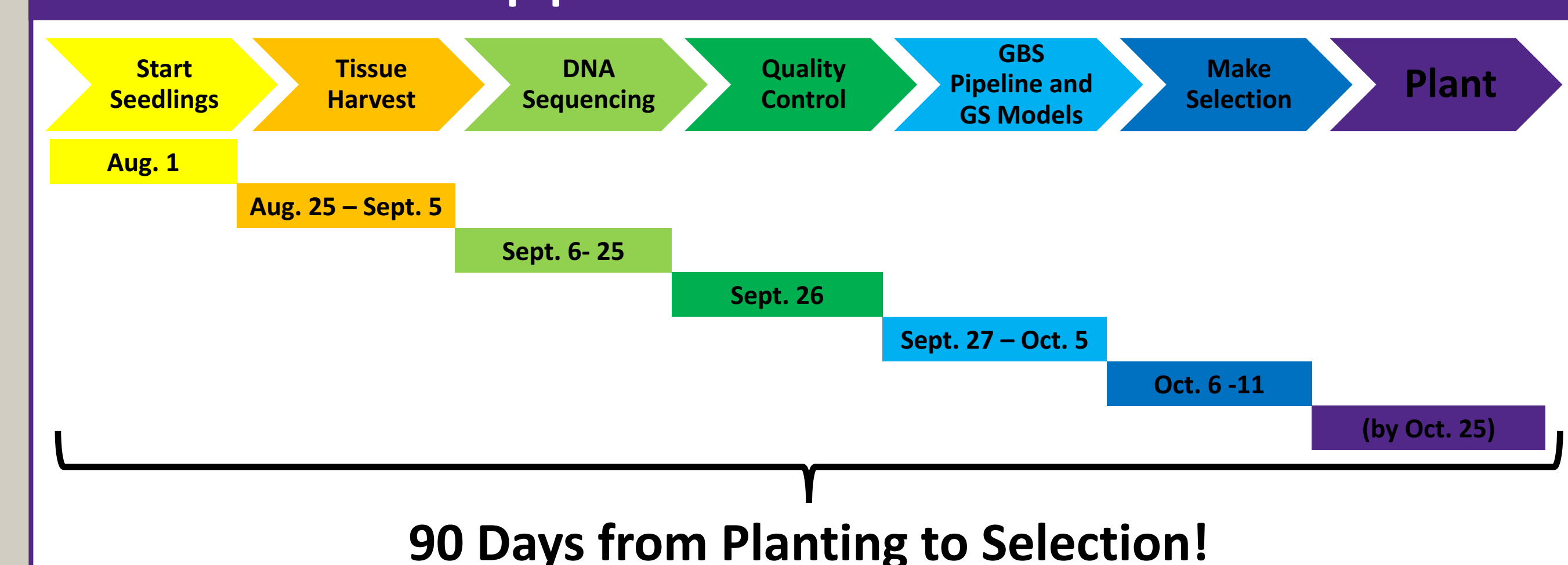

rrBLUP_RR	1.00 ***	1.00 ***	0.96 ***
	BGLR_RR	1.00 ***	0.96 ***
		BGLR_LASSO	0.95 ***
			rrBLUP_GAUS

Figure 3. Comparison of different genomic selection (GS) models for seed weight 2016. The diagonal lists the different GS models including ridge regression, LASSO, and Gaussian kernel using the rrBLUP and BGLR packages. The lower triangle shows the scatterplot results between the models, and the upper triangle displays the correlation coefficient between the different models.

Application Timeline

Start Seedlings — Aug. 1
Tissue Harvest — Aug. 25 – Sept. 5
DNA Sequencing — Sept. 6- 25
Quality Control — Sept. 26
GBS Pipeline and GS Models — Sept. 27 – Oct. 5
Make Selection — Oct. 6 -11
Plant — (by Oct. 25)

90 Days from Planting to Selection!

Refereneces

1. DeHaan, L.R., S. Wang, S. Larson, T. Kantarski, X. Zhang, and D. Cattani. 2014. Current efforts to develop perennial wheat and domesticate *Thinopyrum intermedium* as a perennial grain. In: C. Batello et al., editors, *Perennial crops for food security: Proc. of the FAO expert workshop*. p. 72–89.
2. Zhang, X., Sallam, A., Kantarski, T., DeHaan, L. R., … Anderson, J. A. (2016). Establishment and Optimization of Genomic Selection to Accelerate the Domestication and Improvement of Intermediate Wheatgrass. *The Plant Genome*, 9(1), 1–18.