Assessing Genomic Selection Prediction Accuracy in a Dynamic Barley Breeding Population and Comparing Gain between Genomic and Phenotypic Selection in Barley

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Dedication

I would like to dedicate this work to my wife and my two kids Hussein and Yusuf. I would alike also to dedicate this to my mother and the sole of my father.

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Chapter 1: Assessing Genomic Selection Prediction Accuracy in a Dynamic Barley Breeding Population

Prediction accuracy of genomic selection has been previously evaluated through simulation and cross-validation; however validation based on progeny performance in a plant breeding program has not been investigated thoroughly. We evaluated several prediction models in a dynamic barley breeding population comprised of 647 six-row lines using four traits differing in genetic architecture and 1,536 SNP markers. The breeding lines were divided into six sets designated as one parent set and five consecutive progeny sets comprised of representative samples of breeding lines over a five-year period. We used these data sets to investigate the effect of model and training population composition on prediction accuracy over time. We found little difference in prediction accuracy among the models confirming prior studies that found the simplest model, RR-BLUP, to be accurate across a range of situations. In general, we found that using the parent set was sufficient to predict progeny sets with little to no gain in accuracy from generating larger training populations by combining the parent set with subsequent progeny sets. The prediction accuracy ranged from 0.03 to 0.99 across the four traits and five progeny sets. We explored characteristics of the training and validation populations (marker allele frequency, population structure, and linkage disequilibrium) as well as characteristics of the trait (genetic architecture and heritability). Fixation of markers associated with a trait over time was most clearly associated with reduced prediction

accuracy for the mycotoxin trait DON. Higher trait heritability in the training population and simpler trait architecture were associated with greater prediction accuracy.

Introduction

Genomic selection (GS) is touted as a marker-based breeding approach that complements traditional marker-assisted selection (MAS) and phenotypic selection. In traditional MAS, favorable alleles or genes for relatively simply inherited traits are mapped and then molecular markers linked to those alleles are used to select individuals to use as parents or to advance from segregating breeding populations (Bernardo, 2008). MAS is more effective than phenotypic selection if the tagged loci account for a large portion of the total genetic variation within the population of selection candidates (Collins et al., 2003; Castro et al., 2003; Xu and Crouch, 2008). The limitation of traditional MAS for highly complex traits is that it captures only a small portion of the total genetic variation because it uses a limited number of selected markers (Lande and Thompson, 1990; Bernardo, 2010). Phenotypic selection is effective on quantitative traits, but is limited to stages in breeding cycles and environments where such traits can be measured effectively, such as for advanced lines in multiple location field trials. Therefore, GS can be strategically implemented in breeding for quantitative traits at points in the breeding process where phenotypic selection is not feasible.

Genomic selection uses trait predictions based on estimates of all marker effects distributed across the genome (Meuwissen et al., 2001). Based on simulation studies, genomic selection should improve gain from selection, reduce costs associated with phenotyping, and accelerate development of new cultivars by reducing the length of the breeding cycle (Heffner et al., 2009; 2010). Implementing GS is accomplished by first estimating marker effects in a training population and then using those estimates to

predict the performance of selection candidates. The predicted value of a selection candidate based on marker effects is referred to as the genomic estimated breeding value (GEBV; Meuwissen et al., 2001).

A key component to the effectiveness of GS is prediction accuracy. Prediction accuracy is defined as the correlation between the GEBV and the true breeding value divided by the square root of heritability, which is estimated by measuring phenotypic performance (Goddard and Hayes, 2007; Zhong et al., 2009). There are three general methods to assess prediction accuracy using real data: (i) subset validation, (ii) interset validation, and (iii) progeny validation (Figure 1). *Subset-validation* is implemented by randomly dividing a single population of individuals into equal subsamples; one subsample is used as a validation set to be predicted using the remaining subsamples as the training set. Subset validation has been used to assess prediction accuracy in cattle, wheat, and barley among many other livestock and crop species (Luan et Al., 2009; Heffner et al., 2010; Lorenz et al., 2012; Poland et al., 2012). In *inter-set validation*, predefined sets of genotypes are designated as training and validation populations. These sets could be the same genotypes from independent environments as training and validation data sets or sets of breeding lines chronologically defined where older lines are used to predict newer lines from either the same or independent environments (Asoro et al., 2011; Lorenz et al., 2012). *Progeny validation* implies that the training population includes parents (or grandparents etc…) of progeny lines that comprise the validation population. A simulation study in animals has shown that decreases in prediction accuracy are associated with decay of linkage disequilibrium (LD) between markers and

QTL resulting from recombination in progeny generations (Habier et al., 2007). Therefore meaningful assessment of prediction accuracy should include progeny validation. In plants, we are aware of only a single study that assesses accuracy by progeny validation using empirical phenotypic and genotypic information (Hofheniz et al., 2012).

To assess the potential of GS, researchers have explored various factors that affect prediction accuracy, including prediction models. These models include ridge regression best linear unbiased prediction (RR-BLUP), Bayes A, Bayes B, Bayes Cπ, Bayes LASSO, and Reproducing Kernel Hilbert Space (RKHS) (Meuwissen et al., 2001; Kizilkaya et al., 2010; de los Campos et al., 2009; Gianola and Van Kaam, 2008). These models differ in the assumptions made for marker variances associated with markers and /or types of gene action (reviewed by Lorenz et al., 2011). RR-BLUP assumes that all markers have equal variance whereas Bayes A, Bayes B, Bayes Cπ, and Bayes Lasso models do not impose this constraint (Meuwissen et al., 2001; de los Campos et al., 2009; Kizilkaya et al., 2010). The RKHS regression model can capture both the additive and non-additive interactions among loci by creating a kernel matrix that includes interactions among marker covariates (Gianola and Van Kaam, 2008). Results of empirical studies have shown variable performance of prediction models on different traits (Crossa et al., 2010; Lorenz et al., 2012; Rutkoski et al., 2012).

Other factors shown to affect prediction accuracy include: i) the LD between markers and QTL in the training and the validation populations, ii) the size of the training population (N) , iii) the heritability of the trait under investigation (H^2), and iv) the

genetic architecture of the trait. Increasing marker density will improve prediction accuracy by increasing the number of QTL that are in LD with markers and capturing more of the genetic variation (de Roos et al., 2009; Asoro et al., 2011; Heffner et al., 2011; Zhao et al., 2012). The successful application of genomic selection across generations relies on the persistence of LD phase between markers and QTL (de Roos et al., 2008). The persistence of LD phase measured by the correlation of *r* among populations is likely to be a function of the genetic relationship between populations (de Roos et al., 2008; Toosi et al., 2010). Increasing N will lead to better estimation of SNP effects (Hayes et al., 2009) and therefore, increases prediction accuracy (Lorenzana and Bernardo, 2009; Asoro et al., 2011; Lorenze et al., 2012). In a simulation study, Daetwyler et al. (2010) found that prediction accuracies increased with increase in H^2 of the trait regardless of the number of QTL controlling the trait or the prediction model used. In a study that manipulated H^2 by introducing random error into empirical data sets Combs and Bernardo (2013) showed that accuracy increased with increasing H^2 and N and that prediction accuracies were similar for different combinations when H^2 * N were held constant. Generally, prediction accuracy decreases with the increase of trait complexity (Hayes et al., 2010). Prediction models can vary in performance among traits with different genetic architecture. Bayes B was more accurate when a smaller number of loci control the trait whereas RR-BLUP was insensitive to genetic architecture (Daetwyler et al., 2010).

Previous studies have demonstrated the potential of genomic selection on the basis of subset validation and inter-set validation. While these results are promising,

additional research is needed to assess accuracy in the context of applied breeding. Specifically, validation experiments are needed to assess the accuracy of prediction on progenies (progeny validation) over time as would occur in breeding populations. This would take into account changes in allele frequency and linkage disequilibrium that would be expected to occur as a result of recombination and selection within a dynamic breeding program. Lorenz et al. (2012) investigated prediction accuracy for the disease Fusarium head blight (FHB) and its associated mycotoxin deoxynivalenol (DON) using *interset* validation. In this study, we advance this work by using progeny validation and include additional agronomic traits. We use a set of breeding lines as a training population that include parents that were used to predict five chronological sets of progenies (2006 – 2010) from a breeding program. Our specific objectives were to 1) compare the accuracy of different GS prediction models on DON concentration, FHB resistance, yield, and plant height, 2) study the effect of trait architecture on prediction accuracy, 3) characterize changes in prediction accuracy over time, 4) examine the relationship between prediction accuracy and training population size and composition, allele frequency, linkage disequilibrium, and genetic distance between the training and validation populations.

Materials and Methods

Germplasm

To explore the accuracy of genomic predictions, we utilized historical sets of breeding lines that we define as parent or progeny sets from the University of Minnesota

barley breeding program. The parent set is comprised of 168 breeding lines that were developed between 1999 - 2004 and were either used as parents to develop lines in the progeny sets or were cohorts of breeding lines that were used as parents. The five progeny sets consist of five chronological sets of breeding lines evaluated between 2006 and 2010. Each progeny set consists of approximately 96 lines that were representative of the breeding lines developed that year in the breeding program. The progeny sets 2006 and 2007 are the breeding lines from the University of Minnesota barley breeding program that were included in the association mapping study conducted by Massman et al. (2011) and were referred to as CAP I and CAP II in that study. All the breeding lines in the parental and progeny sets were developed by single seed decent to at least at the F_4. At that point $F_{4:5}$ lines were evaluated for resistance to FHB resistance, heading date, plant height, maturity, and lodging. Lines selected as favorable for these traits are then advanced to preliminary yield trials the following year (Smith et al., 2013). The preliminary yield trial data were used to characterize progeny set lines and the year designation for the progeny set refers to the year that the breeding line entered preliminary yield trials. All pedigree, SNP marker, and phenotypic data related to these sets of breeding lines are available from the public database The Hordeum Toolbox (http://thehordeumtoolbox.org; Blake et al., 2012).

Phenotypic Evaluation

The parental lines were evaluated together for agronomic traits in five experiments conducted between 2009 and 2011 at Crookston and St. Paul, MN in an

augmented block design with two replications and four incomplete blocks per replication (Supplementary Table 1). Planting density for all traits in all experiments was 300 plants/m^2. Each line was represented once per block in two-row plots 3 m in length. Six check cultivars (Drummond, Lacey, Quest, Rasmusson, Stellar, and Tradition) were randomly assigned to each block (Horsley et al., 2002; 2006; Rasmusson et al., 2001; Smith et al., 2010; 2013). We also characterized the parental lines using the historical data that was collected as part of the breeding program as these lines were entered into preliminary yield trials. Experiments for this unbalanced data set were arranged as a randomized complete block design with two replications in two-row plots 3 m in length and were conducted between 1999 and 2004. Three checks (Robust, Stander, and Lacey) were common to all the experiments (Rasmusson and Wilcoxin, 1983; Rasmusson et al., 1993). For both the historic and contemporary data sets, each line was evaluated at least two times in yield trials conducted in St. Paul, Morris, and Crookston, MN. Yield was determined by harvesting each plot with a Wintersteiger small plot combine, weighing the grain, and expressing it as kg/ha. Plant height was assessed as the height in cm of two randomly selected samples of plants from the middle of the plot from the soil surface to the tip of the spike excluding awns. The parental lines were evaluated for FHB resistance and deoxynivalenol (DON) concentration in 2009 at St. Paul and in 2010 at St. Paul and Crookston, MN in an augmented block design with two replications in four incomplete blocks. Each line was represented one time per block in single-row plots 1.8 m in length with 30 cm between rows. Six check cultivars (Drummond, Lacey, Quest, Rasmusson, Stellar, and Tradition) were randomly assigned to each block. The parental lines were

evaluated for FHB resistance and deoxynivalenol (DON) concentration using a previously described method (Steffenson, 2003). Briefly, in St. Paul plants were spray inoculated with a *F. graminearum* macroconidia suspension using CO_2-pressure backpack sprayers. Plots were inoculated when at least 90% of the heads had emerged from the boot and sprayed again three days later (Mesfin et al., 2003). Mist irrigation was applied immediately after inoculation to promote disease infection. In Crookston MN, plants were inoculated by grain spawn using autoclaved corn colonized by five local isolates of *F. graminearum* (Horsley et al., 2006). The colonized grain was spread on the ground two weeks before flowering and again one week later. Overhead mist irrigation started two weeks before anthesis and continued until the hard dough stage of maturity. FHB severity was assessed about 14 days after inoculation by estimating the percentage of infected kernels on a random sample of 10 spikes per plot using the following assessment scale 0, 1, 3, 5, 10, 15, 25, 35, 50, 75, and 100%. DON concentration was determined on a 25g sample from the harvested grain by gas chromatography and mass spectrometry and expressed in parts per million according to the procedures of Mirocha et al., (1998).

Lines included in the progeny sets were derived from crosses made between 2003 and 2007 and were evaluated in preliminary yield trials conducted from 2006 to 2010 (Supplementary Table 1). Plots were arranged in a randomized complete block design with two replications and four check varieties (Robust, Stander, MNBrite, and Lacey). Each progeny set was evaluated for yield and plant height in Crookston, St. Paul, and Morris in MN as described above. The progeny sets were also evaluated for FHB

resistance and DON accumulation in disease nurseries as described above. Each progeny set was evaluated in three to four FHB experiments located in St. Paul and Crookston in MN and Osnabrock and Fargo in ND. Disease inoculation, disease assessment and DON measurements were done as previously described.

Genotypic Evaluation

DNA for genotyping was extracted from a single plant from the $F_{4:5}$ bulk seed used in the phenotypic evaluation. Approximately three week old leaf tissue was harvested and freeze-dried. DNA was extracted at the USDA genotyping center in Fargo, ND using the protocol of Slotta et al. (2008). Each DNA sample was genotyped with the 1,536 SNPs referred to as BOPA1 using the Illumnina GoldenGate oligonucleotide assay (Close et al., 2009). Markers were filtered in parents set based on MAF < 0.01 and missing data frequency greater than 10 percent. Missing marker values were imputed using naïve imputation so that analytical operations could be performed.

Data Analysis

Analysis of variance was performed for DON concentration, FHB resistance, yield, and plant height using the PROC GLM procedure in SAS (9.3) (SAS Institute Inc., 2011). For each experiment, outlier observations with standardized residual absolute values of three or more were removed from the data set and scored as missing values. One experiment (yield in St. Paul, 2010) was removed because no significant differences were found among lines.

To avoid including common checks across experiments in variance component estimates, two-step procedures were used. For the contemporary data from the parental set, we first adjusted phenotypes for block effects by using the common checks among blocks using the PROC MIXED procedure in SAS (9.3) (SAS Institute Inc., 2011). The model was $y = X\beta + Zu + e$ where **y** is the vector of unadjusted phenotypes, **β** is the vector of fixed block effects, and **u** is the vector of random check effects. **X** and **Z** are incidence matrices to relate the vector of unadjusted phenotypes to **β** and **u**. We then adjusted phenotypes for trial effects by estimating these effects as fixed in an analysis with lines as random effects. The model was $y^* = X\beta + Zu + e$ where $\mathbf{y}^*$ are the phenotypes adjusted for block effects calculated in the first step, **β** is the vector of fixed trial effects, and **u** is the vector of random line effects. In the historic data for the parent set, subsets of lines were evaluated in different years, but a common set of checks was included in each trial. Similarly to the contemporary data, phenotypes were adjusted for trial effects by computing these effects in a mixed model with checks as random and trials as fixed effects. In the progeny data sets, phenotypes were adjusted for trial effects by computing these effects in a mixed model with lines as random and trials as fixed effects. Finally, BLUEs for lines in each experiment were estimated in models with adjusted phenotypes as the response variable and lines as fixed effects. Variance components were estimated using restricted maximum likelihood (REML) in the PROC MIXED procedure in SAS by using the line BLUEs as the response variable, lines as random effects and experiments as fixed effects. Broad-sense heritability on an entry mean was estimated for all traits using the equation $H^2 = \sigma_g^2/(\sigma_g^2 + \sigma_e^2/n)$; where σ_g^2 is

genetic variance, σ_e^2 is the pooled error variance that includes GxE and residuals, and *n* is the number of trials.

Characterizing LD, Genetic Distance, and Parental Contribution

To assess the extent of the LD within the parental and progeny sets, the adjacent marker LD was characterized as r^2 using Haploview v4.0 (Barrett et al., 2005). To assess the persistence of LD phase between the parental and progeny sets, the correlations of r were calculated between parental and each progeny set (de Roos et al., 2008; Toosi et al., 2010). We measured genetic distance between the parent set and each progeny set by the fixation index (Fst, Weir and Cockerham, 1984) and Nei's genetic distance (Nei, 1987). Fst and Nei's genetic distances measure the differentiation between two populations due to changes in allele frequencies among populations. The contribution of the parental lines to a progeny set was assessed by summing the number of parents for a progeny line that were included in the parent set over the progeny set and dividing that by twice the number of lines in that progeny set.

Association Analysis

To identify sets of markers associated with traits, association analysis was implemented using the efficient mixed-model association (EMMA) approach, which corrects for population structure using genetic relatedness (Kang et al., 2008). Association analyses were done on the parent set for DON concentration, FHB resistance,

yield, and plant height using *EMMA* package implemented in R (Kang et al., 2008). The analysis was based on the mixed model:

$$y = X\beta + Zu + e \qquad [Eq.1]$$

where $\boldsymbol{y}$ is the vector of individual phenotypes, $\boldsymbol{X}$ is an incidence matrix that relates $\boldsymbol{\beta}$ to $\boldsymbol{y}$, $\boldsymbol{\beta}$ is the vector of fixed effects that includes the overall mean and SNPs, **Z** is the matrix of random effects that relates $\boldsymbol{u}$ to individual phenotypes, $\boldsymbol{u}$ is the random effect of the genetic background of each line and is distributed as $\boldsymbol{u} \sim N(0, \mathbf{K}\sigma^2_g)$. **K** is the kinship matrix derived from marker genotypes and σ^2_g is the genetic variance. $\boldsymbol{e}$ is the residual where $\boldsymbol{e} \sim N(0, \sigma^2_e\ \mathbf{I})$. **I** is the identity matrix and σ^2_e is the error variance (Kang et al., 2008). We used a relaxed threshold of –log *p*-value of 1.3 (*p*-value of 0.05) to identify markers potentially associated with traits. These subsets of markers were used to investigate the changes in allele frequencies over time in the progeny sets. For all polymorphic SNP markers, the proportion of variance explained by each marker (R^2) was calculated as $R^2 = \frac{SS_{reg}}{SS_{Tot}}$, whereas SS_{reg} is the regression sum of squares and SS_{Tot} is the total sum of squares of the regression model.

Prediction Models

Genomic predictions were estimated using four methods: ridge regression best linear unbiased prediction (RR-BLUP; Meuwissen et al., 2001), Gaussian kernel model (Gianola and van Kaam, 2008; Endelman, 2011), Exponential kernel model (Piepho 2009; Endelman, 2011), and Bayes Cπ (Kizilkaya et al., 2010). RR-BLUP and Bayes Cπ can be modeled as

$$y = 1u + \sum_{\{j=1\}}^{K} Z_j \, a_j \delta_j + e \qquad [Eq. 2]$$

where $\boldsymbol{y}$ is the vector of individual phenotypes, u is the population mean, K is the number of markers, $\boldsymbol{Z}$ is the incidence matrix that links marker j genotypes to individuals, $\boldsymbol{a}$ is the effect of marker j, $\boldsymbol{\delta}$ is an indicator variable that indicates the absence or the presence of marker j with probability of π and 1-π respectively, and $\boldsymbol{e}$ is the random residual. In RR-BLUP, all markers are included ($\boldsymbol{\delta}$ = 1) and their effects are distributed with the same variance $N(0, \sigma^2_a)$. The variance of this distribution was estimated on the basis of marker and phenotypic data using restricted maximum likelihood (REML). A Bayesian model was used to relax the assumption of RR-BLUP to allow some marker variances to be zero. Bayes Cπ assumes common marker variance across all markers included in the model; however it allows some markers to have no effect on the trait (Kizilkaya et al., 2010). In Bayes Cπ, it is assumed that each marker j has a zero effect with probability π when $\delta_j = 0$ and an effect $a_j \sim N(0, \sigma^2_a)$ with probability (1- π) when $\delta_j = 1$. The parameter π is treated as unknown and is estimated from the training data. In the Markov Chain Monte Carlo (MCMC) algorithm for Bayes Cπ, 10,000 iterations of Gibbs sampling were used and the first 2,500 iterations were discarded as burn-in. We implemented Bayes Cπ analysis in R (R Development Core Team, 2012). Gaussian and Exponential kernel models were implemented to capture both the additive and non-additive interactions between marker genotypes using the R package rrBLUP (Endelman, 2011; R Development Core Team, 2012). These models can be presented as

$$y = 1u + Zg + e \qquad [Eq.3]$$

where y is the vector of individual phenotypes, u is the population mean, $\boldsymbol{Z}$ is the matrix of that relates $\boldsymbol{g}$ to the adjusted phenotypes, $\boldsymbol{g}$ is the vector of genotypic values that is distributed as $\boldsymbol{g} \sim N(0, K\sigma^2_g)$ where K is the kernel similarity matrix, and $\boldsymbol{e}$ is the residual (Endelman, 2011). The Gaussian and Exponential models do not partition the total genetic variance into additive and non-additive variances; rather, kernel functions are used to capture these effects. Genomic predictions were calculated for all the lines in the validation population using the four prediction models. The correlation coefficient between the genomic predictions and line BLUEs was used to calculate the predictive ability (r_a). Prediction accuracy (r_a/H) of GS (Legarra et al., 2008; Chen et al., 2011) was calculated by dividing the predictive ability by the square root of the broad-sense heritability derived from the validation population data.

Training Populations

To test the effect of training population composition on prediction accuracy, three different scenarios were implemented by varying the training data set. In the first scenario, the 168 parental lines, using either the contemporary or historic data, were used as the training set to predict the performance of lines in each of the five progeny sets. In the second scenario, we varied the training population composition by adding one or more of the progeny sets to the contemporary parent set to predict the performance of a later progeny set. In the third scenario, we used two earlier progeny sets to predict a later

progeny set. For each scenario, we implemented the four prediction models described previously.

Because the experiments described above were used to assess different types of training populations that varied in population size, we also tested the effect of training population size on prediction accuracy for two out of the four traits in 2008 and 2010 progeny sets as validation populations. For DON concentration and yield, we used three scenarios. In the first scenario we randomly sampled 25, 50, 75, 100, and 150 lines from the parent set (n = 168). For each population size, samples were drawn without replacement 500 times. In the second scenario, we combined progeny sets prior to the validation set (combined 2006 to 2007 when predicting 2008 and 2006 to 2009 when predicting 2010) with the parent set into a single panel from which samples were drawn to generate various training sets. We generated training sets from the larger training panels by randomly sampling 25, 50, 75, 100, 150, 168, 264, and 360 when predicting 2008 and sampling 25, 50, 75, 100, 150, 168, 264, 360, and 456 lines when predicting 2010. For each population size, samples were drawn without replacement 500 times. In the third scenario, we combined the parental and progeny sets by sequentially adding each progeny set in chronological order to the parent set such that in each round of prediction the size of the training population was increased by 96. This represents the single case that would occur if a breeder accumulated data over time to increase the size of the training population and thus there exists just one instance for this scenario. For each of the scenarios, we used the training populations to generate predictions of the 2008 and 2010 progeny sets for DON concentration and yield using RR-BLUP.

Results

Phenotypic Traits and Marker Density

The parent set and each of the progeny sets were evaluated in multiple yield and disease experiments between 2006 and 2011. The yield experiment for the parent set in St. Paul 2010 was removed due to very severe lodging that resulted in large error variance and no significant differences among lines. For all traits and experiments, we observed significant differences among lines (p-value < 0.01) in the parent and progeny sets. Genetic variances (Table 1) decreased for DON concentration and plant height as a function of progeny set year, whereas the genetic variances for yield and FHB resistance fluctuated. The estimates of the heritability were moderate for DON concentration and FHB resistance; low to moderate for yield; and high for plant height as expected based on previous studies (Boukerrou and Rasmusson, 1990; Ma et al., 2000; Mesfin et al., 2003). After filtering the 1,536 BOPA1 SNPs for MAF and missing data, 984 markers remained that spanned 1085 cM of the barley genome with an average distance between adjacent markers of 1.1 cM.

Relationship between Parent and Progeny Sets

The average adjacent marker LD in the progeny sets were greater than the parent set and showed a slight increase over time (Figure 2). The correlation of r between parental and progeny sets ranged from 0.44 to 0.61 (Figure 2). The parental contribution of the parent set to the progeny sets decreased continuously over time with about a 75% reduction from 2006 to 2010 (Figure 3). Concurrent with this decrease in parental

contribution was an increase in genetic distance between parent and the consecutive progeny sets over time (Figure 3). The genetic relationship between the parents and the progeny sets can be visualized in the heatmap of the kinship matrix (Figure 4). As lines were developed in the breeding program, their similarity to the parent set diminished over time.

Marker-Trait Associations

Based on association analysis using the parent set, all of the traits displayed quantitative inheritance with multiple loci distributed across the genome contributing to the traits (Figure 5). Coincident QTLs for plant height, DON concentration, and FHB resistance on the short arm of chromosome 4H were detected in a region previously identified in a study using a similar germplasm (Massman et al., 2011). Using a relaxed *p*-value of 0.05, we identified 62, 58, 62, and 59 markers associated with DON concentration, FHB resistance, yield, and plant height, respectively.

To characterize the possible role of selection, we examined allele frequencies of the SNP markers associated with the four traits (Figure 6). In general, there was an increase over time with the complete set of genome-wide markers. For markers associated with individual traits this trend was most apparent for DON concentration followed by plant height and yield. No relationship was observed for FHB resistance.

To investigate the effect of trait architecture and the distribution of maker effects on prediction accuracy, we estimated the proportion of variance explained by each SNP marker in the parent set for all traits (Figure 7). Based on the distribution of R^2 values, we

found that plant height was the least complex trait with several markers exceeding 0.30. Yield, on the other hand, was the most complex trait with only a few markers with R^2 values greater than 0.10. Based on Figure 5, it is clear that multiple markers are likely associated with the same QTL. Nevertheless, 83, 59, 24, and 17 markers had R^2 values greater than 0.10 for plant height, DON concentration, FHB resistance, and yield respectively.

Another way to characterize the genetic architecture of the traits is to use the π parameter, which is the proportion of markers with no effect, estimated from the Bayes Cπ modeling. When using the parent set as a training population, the π parameter estimates for yield over 4 runs ranged between 0.28 – 0.43 with a mean π of 0.35. For DON concentration the π parameter estimates ranged between 0.37 – 0.54 with a mean π of 0.45. For FHB the π parameter estimates ranged between 0.49 – 0.58 with a mean π of 0.53. For plant height the π parameter estimates ranged between 0.45 – 0.80 with a mean π of 0.63. Thus, based on π estimates, yield was the most complex trait followed by DON concentration, FHB resistance, and then plant height. This suggestive trend from higher to lower complexity agrees with the distribution of R^2 values for markers displayed in Figure 7. Assessment of genetic architecture based on the π parameter estimates also agrees with the results of (Lorenz et al., 2012) for DON concentration and FHB resistance.

Prediction Accuracy

For the four traits investigated, all prediction models performed similarly to each other with respect to prediction accuracy (Supplemental Figure 1). When we averaged the prediction accuracy across the five progeny set years, we found no significant differences among the models (Supplemental Table 2). There was a strong correlation among the four models for the predictions of yield for the combined set of progeny when using the parent set as a training population (Figure 8). Consistent with other GS studies RR-BLUP, in which all marker effects are sampled from the same distribution and similarly shrunken toward zero equally, performed similarly to models that do not impose that restriction. Further comparisons of prediction accuracy are based on RR-BLUP.

Another important consideration for prediction accuracy is the need to generate new phenotypic data for model training or to use existing data sets. We estimated marker effects using available historical data for yield from the breeding program in the parent set and compared that to estimates obtained using the contemporary data set (Figure 9). The average prediction accuracy over the five years based on contemporary (0.57) and historic data (0.42) were not significantly different (p-value=0.38). Combining historic and contemporary data was equal to using the contemporary data alone.

In general, when using the parent set to predict progeny sets, accuracy was highest for plant height and lowest for yield (Figure 10). FHB resistance and DON concentration had similar prediction accuracy. The relationship between accuracy and year of the progeny set also differed between traits (Figure 10). For yield and plant height

the prediction accuracy fluctuated over time while for DON concentration there was an overall decrease. Accuracy for FHB resistance remained relatively constant across years.

Varying the size of the training population by adding one or more progeny sets to the parent set in order to predict a later progeny set generally showed the same trend observed for the parent set alone and in several instances resulted in reduced accuracy. Using the most recent breeding lines and environments to the test population by training a prediction model from the two progeny sets prior to the validation year was generally less accurate than using the parent set. The general trend was that higher trait heritability in the parent training population corresponded to higher predictive ability when parents predicted all the progenies using RR-BLUP (Figure 11).

Since adding consecutive sets of lines to the parent set changed both the composition and the size of the training set, we looked at the effect of population size on prediction accuracy with the parent set only and with the parent set combined with the progeny sets with different population sizes drawn at random. In both cases, we identified an increase in accuracy with increasing in population size for DON concentration and yield (Figure 12). However, prediction accuracy for DON concentration seemed to plateau at a population size of 75 while yield did not appear to plateau. It was also interesting that random sampling from just the parent set often produced higher prediction accuracies than random sampling from the combined parent and progeny sets when compared at the same training population size.

Discussion

Successful implementation of genomic selection will involve the use of improved genotyping technology to shorten the breeding cycle and increased selection intensity by effective modeling to accurately predict breeding values. Prior studies have examined factors that affect prediction accuracy through simulation and empirically through cross-validation (eg. Daetwyler et al., 2010; Lorenz et al., 2012). To assess prediction accuracy in a more realistic context, we used sets of parents and progenies from an active breeding program as training and validation sets. Because breeding populations are dynamic, we tested progeny sets defined chronologically over a five year period. We found that prediction accuracy varied over time and that simply adding data from breeding progenies to the training population did not improve and often reduced prediction accuracy. This suggests that careful construction of training populations is warranted. We considered the relationship between the training and the validation populations with regard to genetic distance and differences in linkage disequilibrium and allele frequencies. In this breeding population, all of these factors changed over time and each individually could not completely account for differences in prediction accuracy. However, the data support their role in affecting prediction accuracy and suggest they should be taken into account when designing training data sets and developing strategies for retraining models over time to maintain acceptable levels of accuracy.

Prediction Accuracy can be Affected by Changes in Breeding Populations over Time

Breeding populations are dynamic and as such approaches to using prediction methods should be informed by changes in prediction accuracy that may occur over time. Breeding value predictions are influenced by allele frequencies, LD level, and the introgression of new alleles. These factors will change over breeding cycles due to selection, genetic drift, and unequal parental contribution to progenies. We investigated prediction accuracy in validation sets of breeding lines over a five year period and observed both little to no change as well as substantial decrease in prediction accuracy over time depending on the trait. To better understand the underlying population parameters that could be affecting prediction accuracy, we compared the parental training population to the progeny validation sets with respect to allele frequencies, parental contribution, genetic distance, and LD.

More than 35% of marker alleles that were segregating in the parent set became fixed in the 2010 progeny set. Gradual increases in allele fixation for trait specific markers are an indication of the effect of selection and/or genetic drift. A previous study of the University of Minnesota barley breeding program showed that a reduction in allelic diversity for specific simple sequence repeat markers was in some cases associated with QTL regions for traits that were under selection (Condón et al., 2008). Once a marker associated with a trait that is segregating in training population becomes fixed in subsequent progeny generations, it loses its predictive value for the purpose of selection. We observed a substantial increase in the number of fixed SNPs associated with DON that corresponded to a reduction in prediction accuracy. However, we saw a similar

increase in fixed SNPs for yield and no corresponding reduction in accuracy. One possible explanation is that yield is likely conditioned by a greater number of QTL with smaller effects and therefore increases in the number SNPs that become fixed over time would have less of an effect on prediction accuracy.

Prediction accuracy should be greatest when the training population is more closely related to the validation population (Habier et al., 2007; Hayes et al., 2009; Lorenz et al., 2012). We observed in most cases that the 2006 progeny set, which was genetically more similar to and had the largest number of direct parents from the parent set, was predicted with the greatest accuracy. The increase in the genetic distances between the parental and progeny sets was most closely associated with a decline in prediction accuracy for DON but not for FHB resistance, yield, and plant height. This indicates that other factors may also contribute to changes in prediction accuracy over time.

Populations can differ in the degree of the LD between markers and QTL due to drift, selection, and/or recombination (Dekkers, 2004; Barton and Otto, 2005). Prediction accuracy should increase as LD between markers and QTL increases. Recombination in breeding populations should reduce LD between markers and QTL over the time while selection and genetic drift should increase LD (Pfaffelhuber et al., 2008). Habier et al. (2007) studied the effect of LD on prediction accuracy over many generations and found a decrease in prediction accuracy was associated with decay of LD. We found a general increase in adjacent marker LD in the progeny sets over time while prediction accuracy generally remained constant or decreased. We also examined the persistence of adjacent

marker LD between the parent set and each of the progeny sets using the correlation of *r* (de Roos et al., 2008; Toosi et al., 2010). The correlation of *r* did not decay over the window of time of this experiment despite the fact that genetic distance between the parents and progeny sets increased over time. Asoro et al. (2011) suggested that the ability of early generations to predict later generations was due to the persistence of the LD phase between early and late generations. Thus, even if validation populations become more genetically distant from training populations, if the LD phase is consistent, prediction accuracy will be maintained.

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How do Trait and Population Characteristics Affect Prediction Accuracy?

Ideally genomic selection can be applied to traits that vary in heritability and genetic architecture. In our study based on estimates of R^2 and the π parameter, yield was the most complex trait while plant height was the least complex. However, inference of genetic architecture based on π should be interpreted cautiously (Gianola, 2013). We found that yield, a more complex and lower heritability trait, generally had lower prediction accuracy than a simpler and higher heritability trait such as plant height. This is consistent with other studies where complex traits controlled by many loci with small effects produced lower prediction accuracy than less complex traits (Hayes et al., 2010). Genomic predictions should be more accurate for traits with higher heritability (Hayes et al., 2009; Daetwyler et al., 2010; Lorenz, 2013; Combs and Bernardo, 2013). Prediction accuracy for yield in the current study was higher than accuracy observed for yield in oats (Asoro et al., 2011). They reasoned that lower accuracy for oat yield was due to the

evaluation of their germplam in a wider range of environments which reduced the genetic variance relative to the GxE variance and thereby reduced heritability for yield. The barley germplasm in the current study was evaluated in more homogenous environments, which are the target production and evaluation environments in Minnesota. Therefore, the genetic variance is expected to be higher relative to GxE leading to a higher heritability estimate and an increased prediction accuracy.

In addition to the characteristics of trait, heritability, and genetic variance; LD (as discussed above) and population structure can affect prediction accuracy. These factors could have contributed to the striking difference in the response of accuracy to increased training population size for DON versus yield (Figure 12). Both traits have higher than expected accuracies at very low training population sizes (e.g., 25 individuals). Windhausen et al. (2012) suggested that high accuracy at low training population size can be diagnostic of subpopulation structure affecting accuracy. In this context, we suggest that structure could reduce accuracy at high training population size from the following mechanism. Population structure is a cause of LD: two loci that both have differences in allele frequency across subpopulations will be in LD. Thus, structure can cause association between a marker and several QTL. This phenomenon has been an ongoing issue in genome-wide association studies (e.g., Pritchard et al., 2000). In the context of genomic prediction, structure-generated disequilibrium between a marker and several QTL will prevent the marker's estimated effect from converging on the effect of a QTL to which it is actually linked, regardless of the training population size. Though they did not comment on it, Wimmer et al. (2013) observed a phenomenon like this: in their

Figure 6A, at low model complexity, the error of marker effect estimates increases as training population size increases. This increase in error arises presumably because of the documented deep structure in rice (Zhao et al., 2011). The question remains as to why this mechanism would more strongly affect DON than yield. We hypothesize that structure in the Minnesota barley breeding program is more strongly correlated to DON than to yield, given that it has been purposefully split into a population where FHB resistance was prioritized versus one where yield and quality continued to be prioritized (Fang et al., 2013).

The availability of genome-wide markers can improve our understanding of genetic architecture and the extent to which epistasis influences complex traits. In general, the four genomic prediction models tested produced similar accuracies across the four traits investigated in this study. The four models differed in assumptions about the genetic architecture of the trait and the extent to which non-additive effects contribute to the prediction. Lande and Thompson (1990) suggested the use of epistatic effects in addition to additive effects in marker-assisted selection schemes. Liu et al. (2003) found that including epistasis improved both the response and efficiency of marker-assisted selection. In some studies, including epistasis in genomic prediction models through the use of non-additive kernels resulted in increased prediction accuracy over RR-BLUP (Crossa et al., 2010; Wang et al., 2012). In our study, we found that simple additive models (RR-BLUP and Bayes Cπ) performed similarly to those that account for both additive and non-additive effects (Exponential and Gaussian). These results are similar to a recent study of barley breeding lines evaluated for DON concentration and FHB

resistance that showed that both Bayes Cπ and RR-BLUP produced the same level of accuracy (Lorenz et al., 2012).

Practical Implications for Breeding

The increasing ease and rapidly declining cost of genotyping means that assembling phenotype data to train prediction models will be the limiting step to implementing genomic selection. We found that using the contemporary parent data was slightly, but not significantly, better than using historic parent data to train a prediction model. The contemporary data was balanced and we corrected for field spatial variability using the common checks whereas the historic data was unbalanced and no correction for field variability was made. Nevertheless, the prediction accuracy from historic data was in most cases around 0.50 for each of the five years and this level of accuracy suggests genomic selection would be effective if the breeding cycle time is half of what is done in phenotypic selection (Asoro et al., 2011). These results suggests that breeders could reduce time and costs by using unbalanced historical data after proper adjustment for spatial variability and trial effects to train a prediction models. Historical unbalanced phenotypic data were also used to assess the use of genomic selection in oats (Asoro et al., 2011). Initiating genomic selection with existing data and later incorporating contemporary data sets should allow breeders to realize benefits of genomic selection sooner and improve effectiveness over time.

The size and composition of the training population are important factors to manipulate prediction accuracy. Breeders may consider combining training data sets to

maximize the use of the available phenotypic and genotypic information and generate larger population sizes (Hayes et al., 2009; de Roos et al., 2009; Asoro et al., 2011; Lorenz et al., 2012; Technow et al., 2013). Lorenz et al. (2012) found little to no improvement in prediction accuracy for FHB resistance and DON concentration when increasing the size of the training population by combining different barley breeding populations. Conversely for maize, when combining both flint and dent heterotic groups together, prediction accuracies increased by 10% and 13% when predicting dent and flint heterotic groups respectively for northern corn leaf blight resistance (Technow et al., 2013). In our study, we found only a slight improvement or a reduction in accuracy when increasing the population size by adding progeny sets from the same breeding program to the parent set. However, when adding progeny to the parent set, both the size and the composition of the training populations were altered. Therefore, we separated these two factors by generating training populations by randomly sampling from the combined data set. Interestingly, the prediction accuracy for DON concentration plateaued at a much smaller population size compared to yield (Figure 12). Prediction accuracy for yield did not level off suggesting that the benefit from increasing training population size may depend on the trait.

In addition to optimizing prediction accuracy, the effectiveness of genomic selection will increase by shortening the breeding cycle time and reducing the cost of selection (Heffner et al., 2010; Jannink et al., 2010). In the University of Minnesota barley breeding program, genomic selection is implemented at the F_3 stage for FHB resistance, DON concentration, and yield. This is one year after crossing parents

compared to a four year breeding cycle that is typical for phenotypic selection. The prediction accuracies that we observed based on progeny validation always exceeded 0.25 indicating that GS should exceed phenotypic selection in gain per unit time. Combined with rapidly decreasing genotyping costs this suggests that genomic selection should improve breeding efficiency substantially.

Table 1. The estimated genetic (σ_g^2) and error (σ_e^2) variances for the parent contemporary and progeny sets (2006 – 2010) for deoxynivalenol concentration (DON), Fusarium head blight resistance (FHB), yield, and plant height (HT).

	$\boldsymbol{\sigma_g^2}$				$\boldsymbol{\sigma_e^2}$			
Year	**DON**	**FHB**	**Yield**	**HT**	**DON**	**FHB**	**Yield**	**HT**
Parents	15.83	12.00	114411	26.20	20.00	35.00	444284	11.30
2006	51.23	34.80	97701	19.30	122.23	89.60	330349	8.90
2007	15.40	4.60	109203	15.20	24.01	16.00	334950	9.50
2008	23.20	14.40	280128	28.40	74.10	20.20	274314	7.90
2009	12.90	6.95	68516	14.50	22.90	22.99	360820	11.30
2010	7.50	13.99	95241	7.96	16.80	58.60	365080	8.10

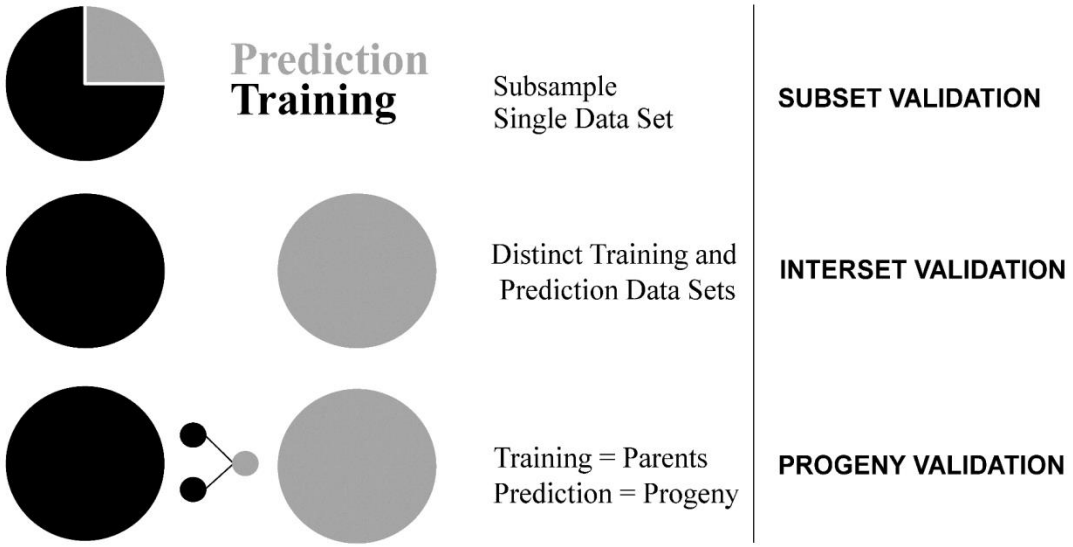

Figure 1. Three validation approaches to assess prediction accuracy using different training and prediction sets.

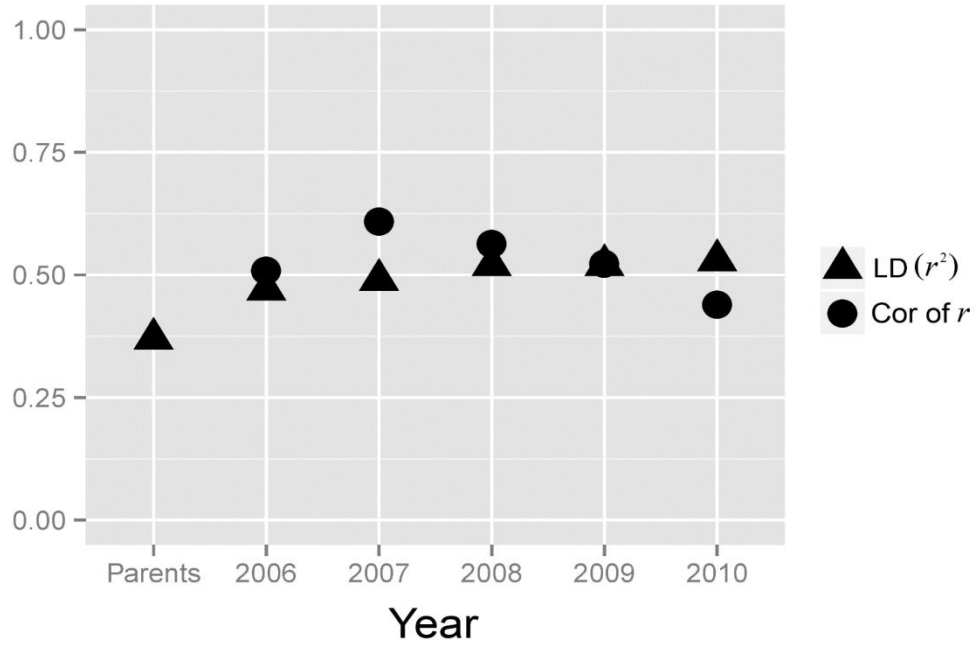

Figure 2. The average linkage disequilibrium (LD) of all possible adjacent marker pairs in the parental and progeny sets (triangle) and the correlation of *r* between parents and each of the progeny sets (circle).

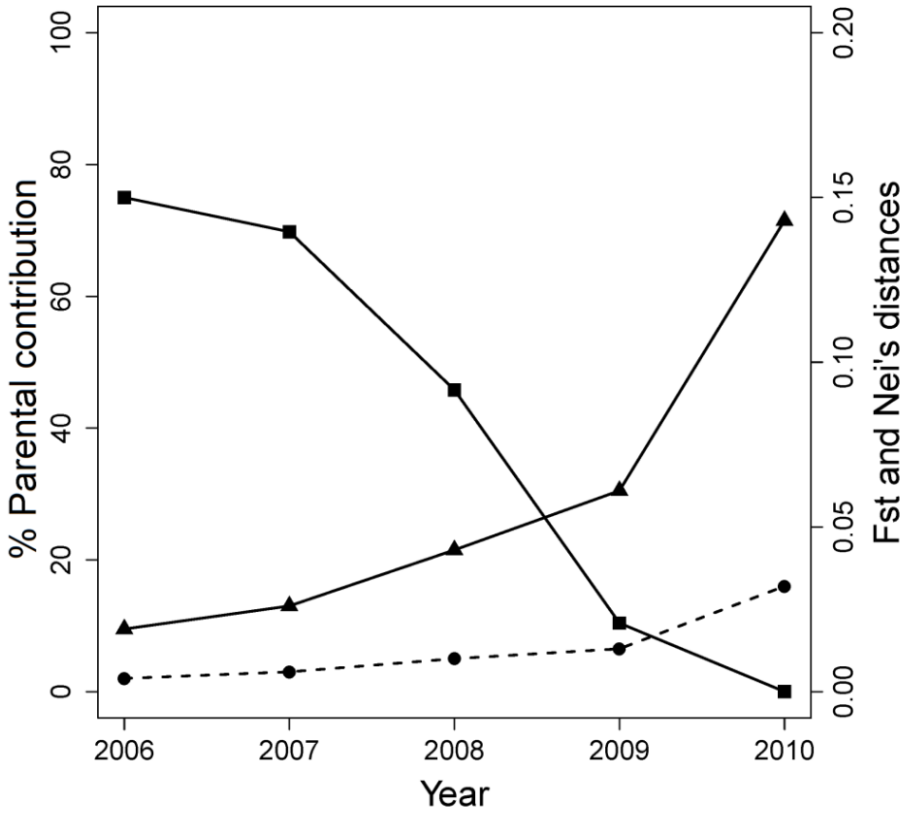

Figure 3. Relationship between the parent set and each progeny set expressed as percentage of parental contribution (square) to each of the progeny sets and the genetic distance between parental and progeny sets expressed as *Fst* (triangle) and *Nei* genetic distances (circle).

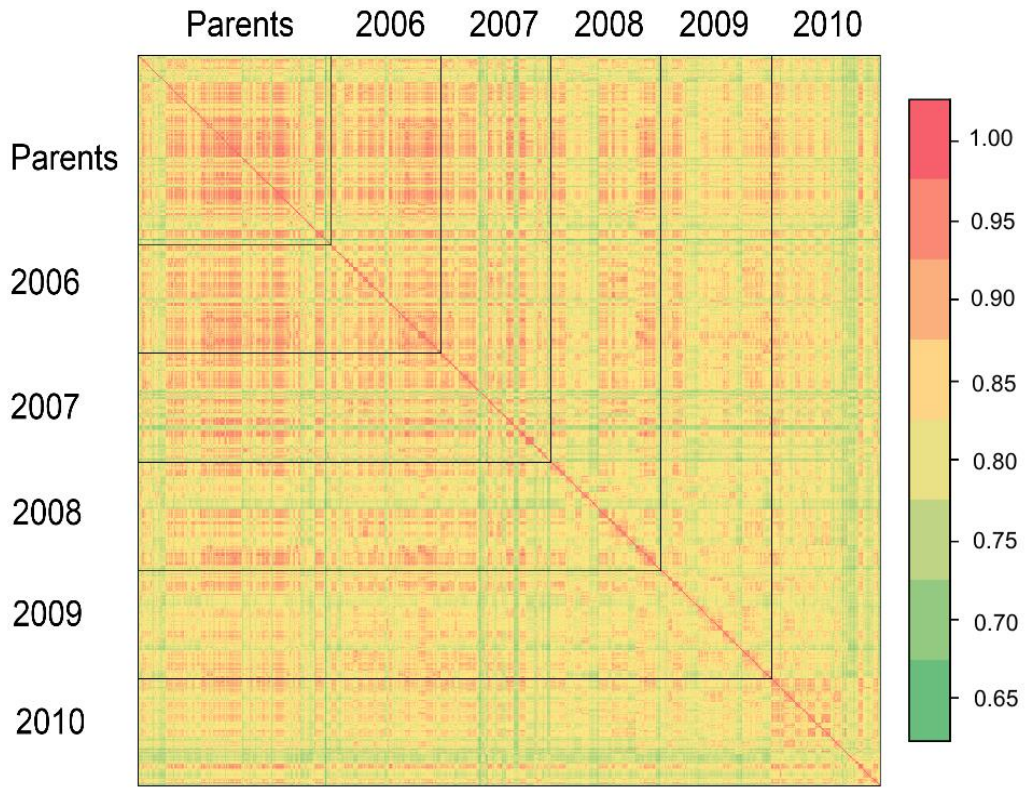

Figure 4. Heatmap displaying the similarity kinship matrix calculated using marker data for parents and all progeny sets.

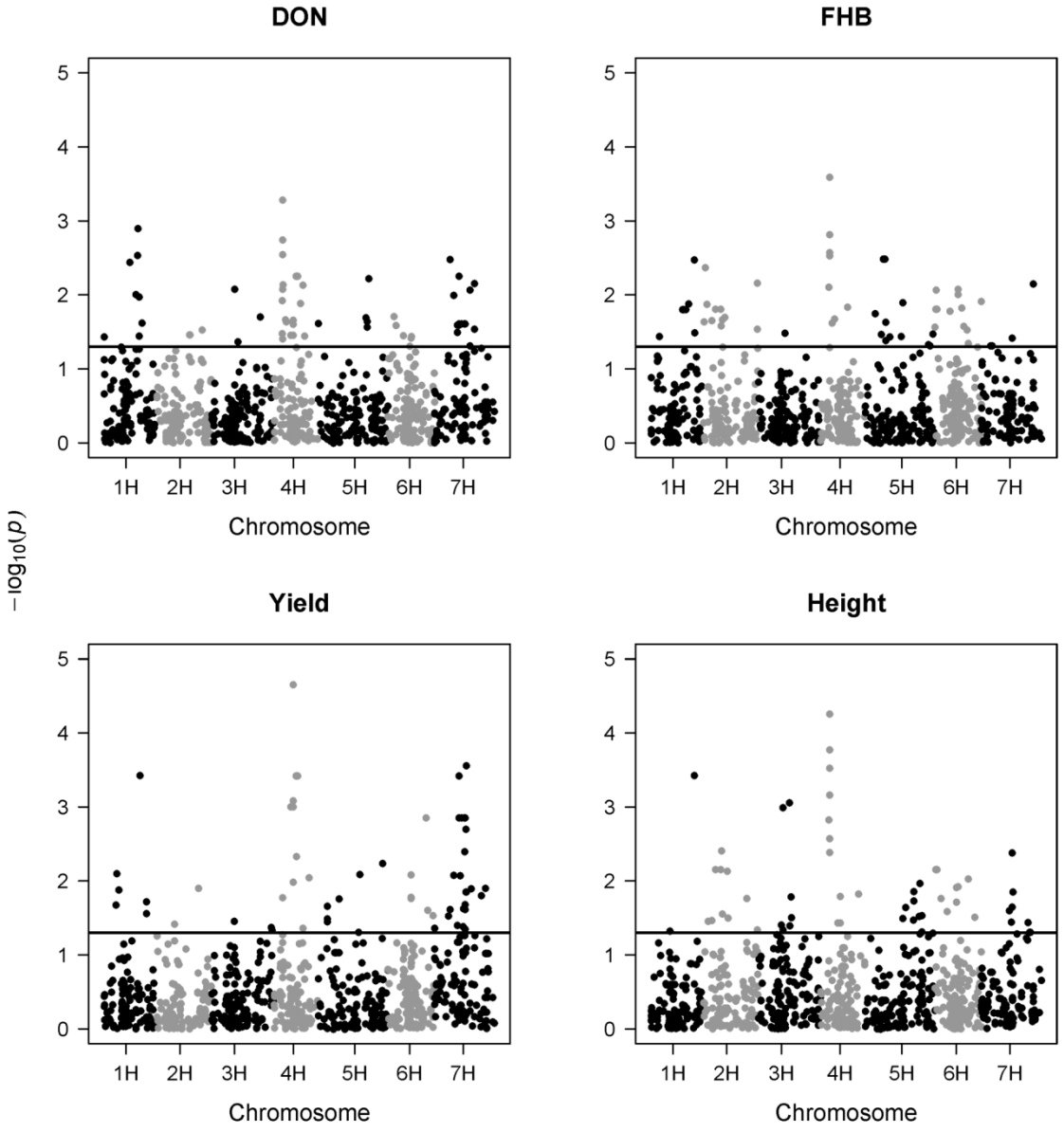

Figure 5. Manhattan plot displaying significance level for association mapping of deoxynivalenol (DON) concentration, Fusarium head blight (FHB) resistance, yield, and plant height in the contemporary parent data set. The relaxed threshold of 1.3 –log(p) which corresponds to *p*-value of 0.05 is shown with a horizontal line.

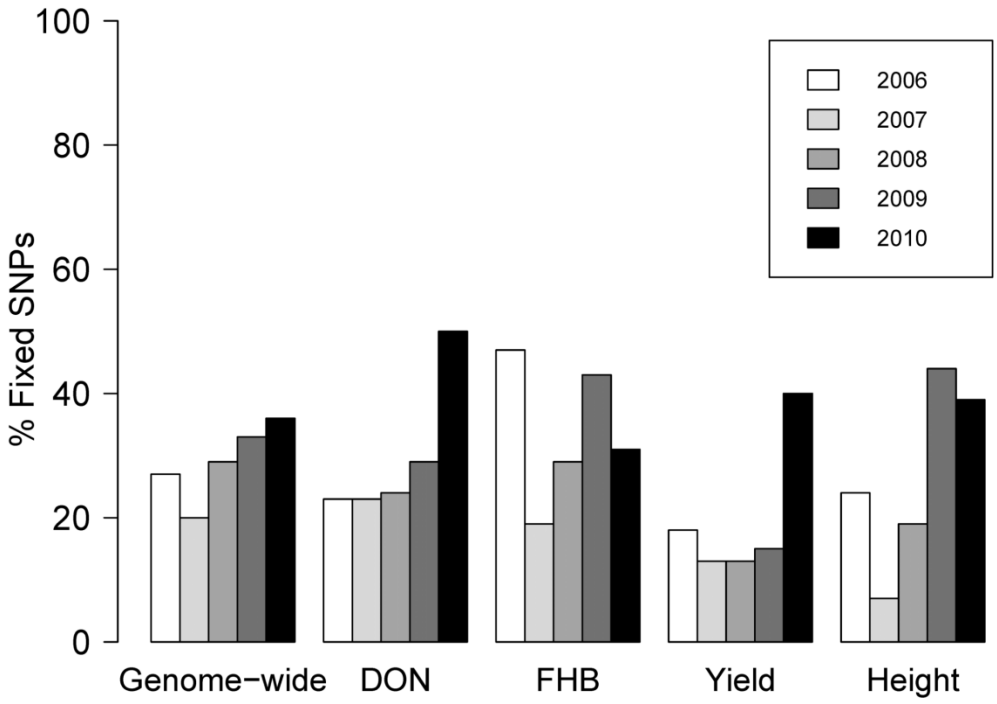

Figure 6. Percentage of SNPs that are fixed in the complete marker set (genome-wide) and in the subsets of markers associated with deoxynivalenol (DON) concentration, Fusarium head blight (FHB) resistance, yield, and plant height (see Figure 5) in each of the five progeny sets between 2006 and 2010.

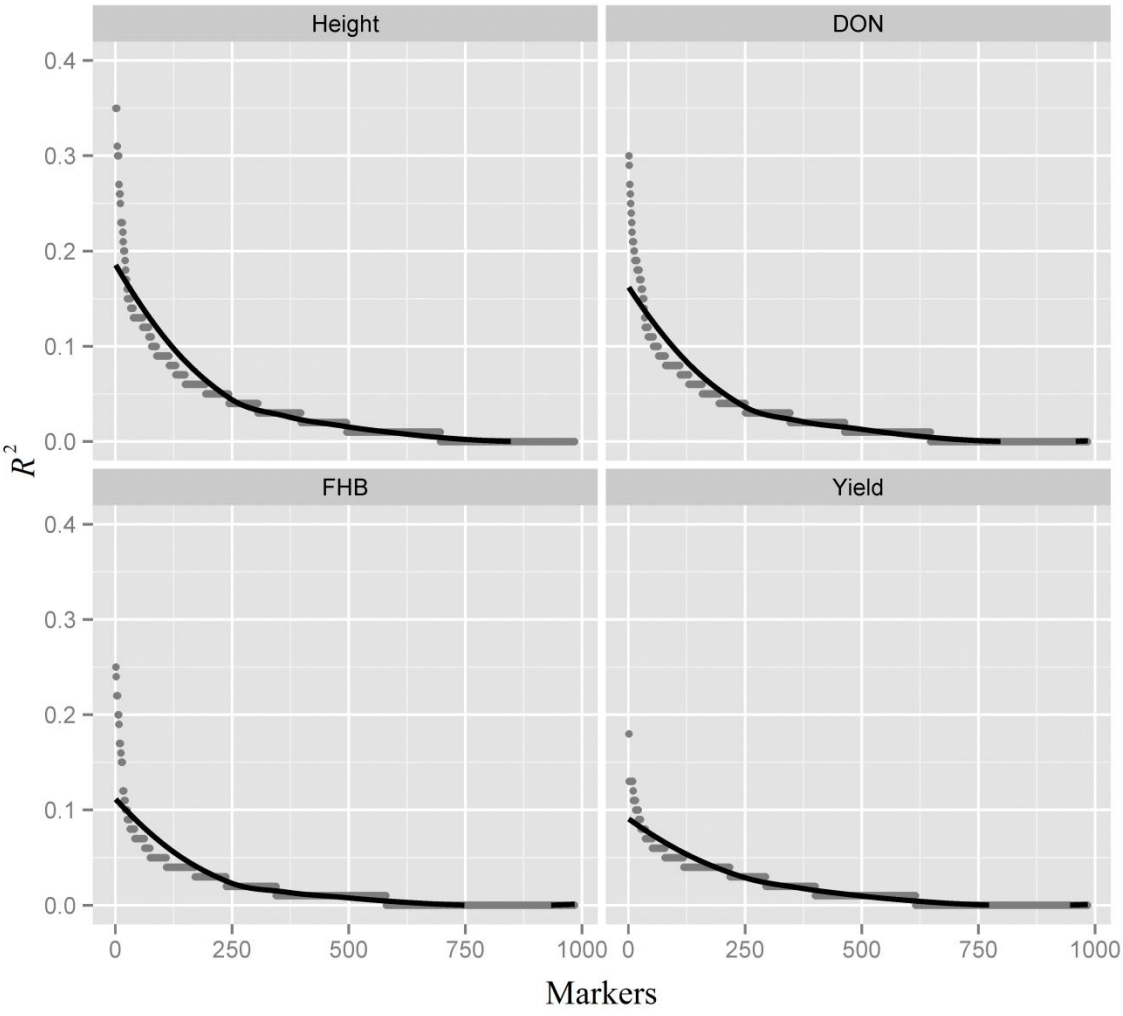

Figure 7. Distribution of marker R^2 values for plant height, deoxynivalenol (DON) concentration, Fusarium head blight (FHB) resistance, and yield. R^2 is the proportion of genetic variance explained by a marker.

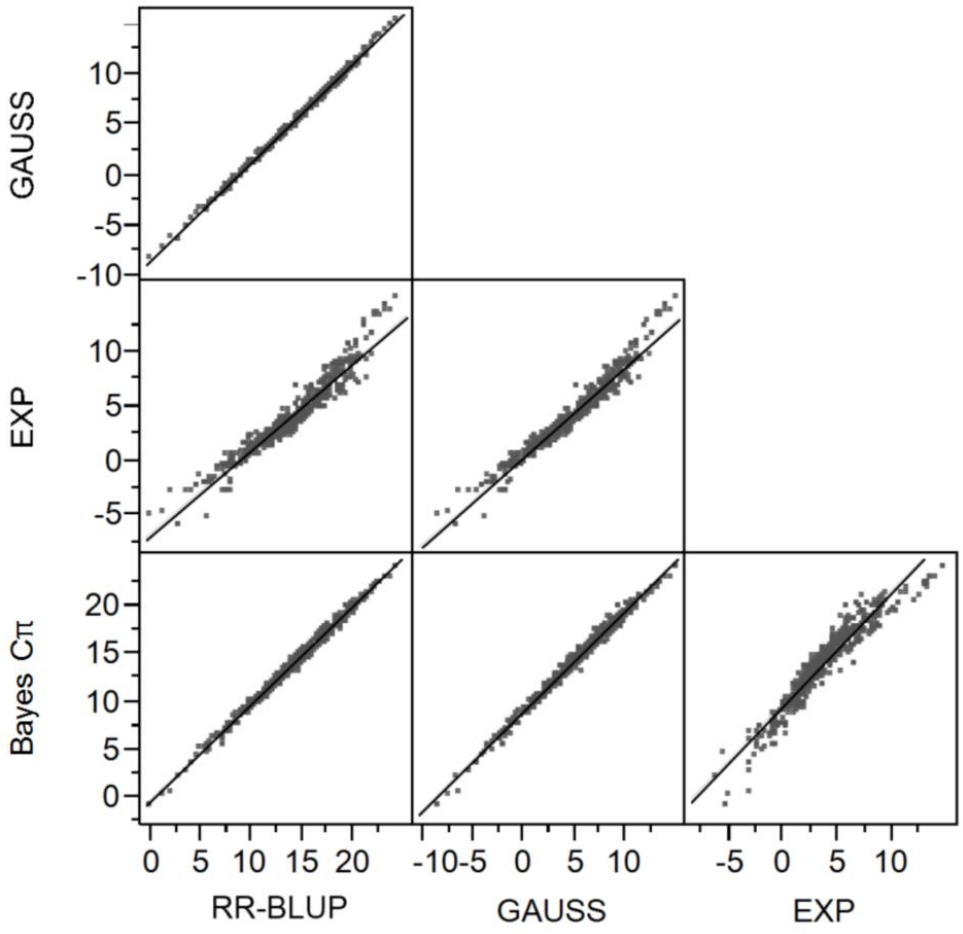

Figure 8. Scatterplot matrix for all prediction models when using contemporary parent data set as the training population to predict all progeny sets (2006-2010) using ridge regression best linear unbiased prediction (RR-BLUP), Gaussian kernel model (GAUSS), Exponential kernel model (EXP), and Bayes Cπ for yield.

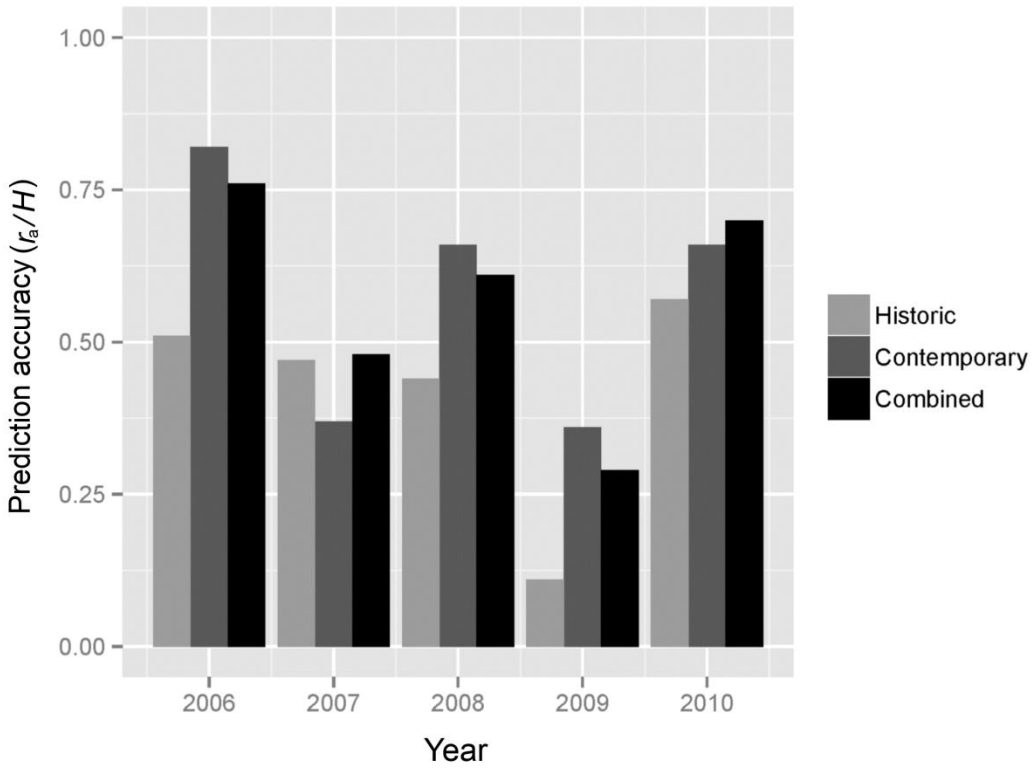

Figure 9. Prediction accuracy for yield using historic, contemporary, and combined (historic and contemporary) parent data to predict five progeny sets using RR-BLUP.

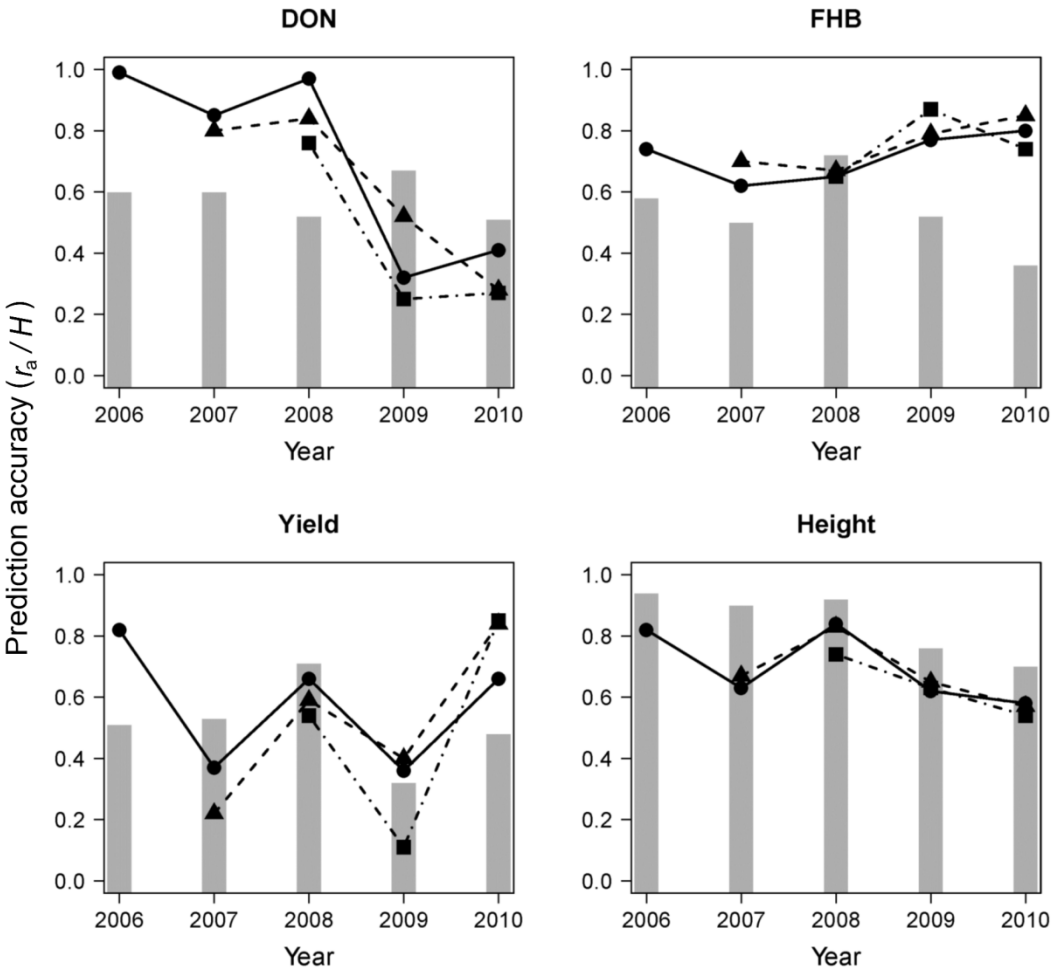

Figure 10. Prediction accuracy for the four traits using RR-BLUP in three scenarios for training populations: using the contemporary parent data set to predict each progeny set (circle), using the sequential addition of progeny sets to the contemporary parent data set to predict the later progeny set (triangle), and using the two previous years of the progeny sets to predict the later progeny set (square). The heritability for each progeny set used as the validation set is shown in the solid bar.

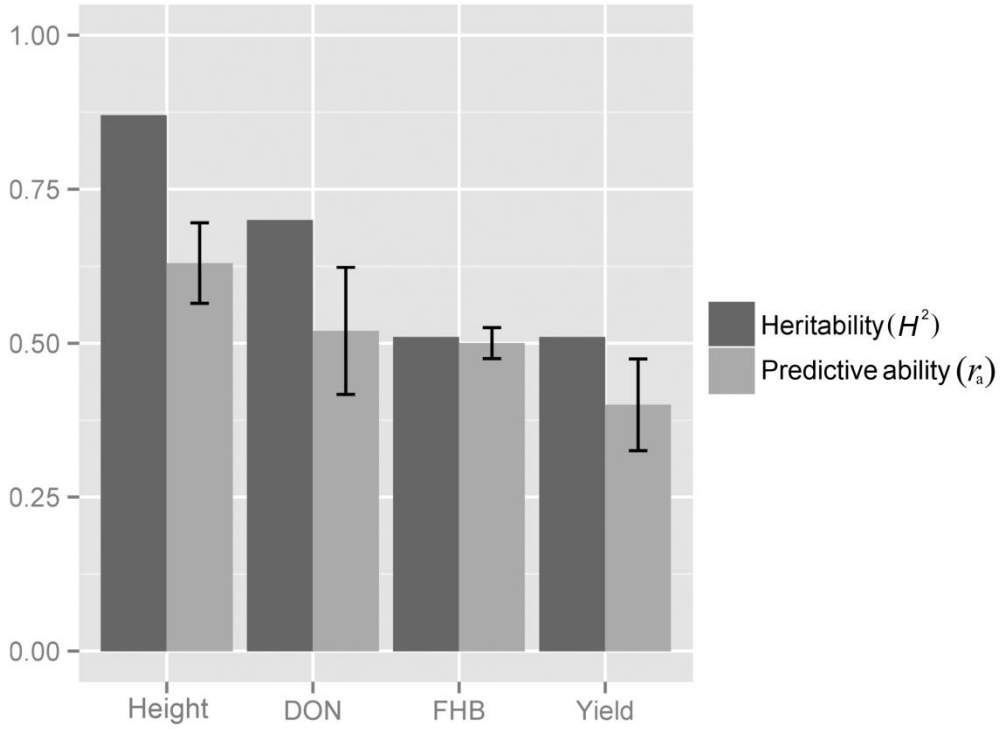

Figure 11. Relationship between the predictive ability (correlation between GEBV and line BLUEs) when using contemporary parent data set to predict progeny sets using RR-BLUP and heritability of the contemporary parent training population for plant height, deoxynivalenol (DON) concentration, Fusarium head blight (FHB) resistance, and yield.

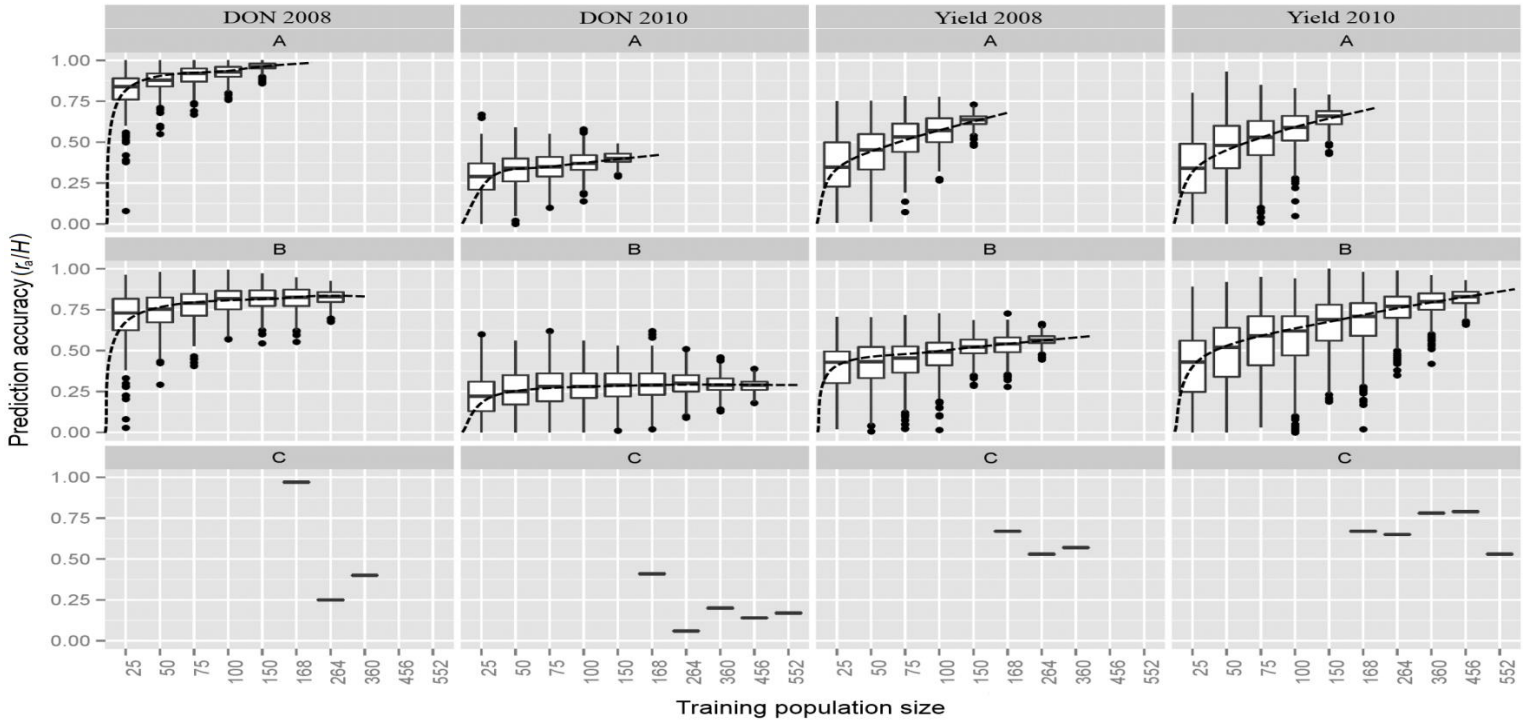

Figure 12. Relationship between population size and prediction accuracy for deoxynivalenol (DON) concentration and yield. Three scenarios are presented:

A) Using the contemporary parent data set as the training population to predict the 2008 and 2010 progeny sets. Each point represents a subset of the training population by random sampling 500 times.

B) Using the combined contemporary parent data set and progeny sets prior to the validation set to predict the 2008 and 2010 sets. Each point represents a subset of the training population by random sampling 500 times

C) The sequential addition of a progeny set to the contemporary parent data set as a training population to predict the 2008 and 2010 progeny sets. The training population sizes are 168 (parent set), 264 parent set + 2006 (n=96), etc…

Chapter 2: Comparing Genomic and Phenotypic Selection in Barley

Genomic selection is a marker based selection method that promises to improve and accelerate the breeding process in plants and animals. Numerous studies have investigated the gain per unit time; however very limited studies have directly compared gains from genomic and phenotypic selection using empirical data. In this study, we used five consecutive sets of breeding lines to compare the gain between genomic and phenotypic selection. In each set, about ninety six barley breeding lines were phenotypically evaluated for yield, FHB resistance, and DON accumulation. A set 168 historic parental lines were used as a training population to predict the performance of the selection candidate sets using RR-BLUP. All lines were genotyped using 1,536 SNP markers (BOPA1) for all seven barley chromosomes. The best performing 10% of the breeding lines in each year were selected using the two schemes and revaluated together in several trials in Minnesota and North Dakota. We made direct comparison between genomic and phenotypic selection in two selection candidate sets for yield and five sets for FHB resistance and DON accumulation. We assessed the relative efficiency of genomic over phenotypic selection and changes in the genetic similarity using the two selection schemes. Results showed similar response to selection between genomic and phenotypic selection in most cases. Genomic selection resulted in more genetic similarity only for FHB resistance; however, for yield and DON concentration no changes in the genetic similarity were detected between genomic and phenotypic selection. In addition, we assessed the use of phenotypic selection for FHB, genomic selection for FHB, and genomic selection for DON as indirect selection methods to select for low DON

concentration. We did not find significant differences between direct and indirect selection methods.

Introduction

The goal of selection is to increase the frequency of beneficial alleles in a breeding population to increase genetic gain over time (Falconer and Mackay, 1996). Improvement of quantitative traits has relied mainly on phenotypic selection (PS) where the value of each line is derived from limited number of phenotypic evaluations across environments. The determination of a line's genotypic value using limited phenotypic evaluations suffers from low accuracy for traits with low heritability. Effective selection requires an accurate estimation of breeding values to identify breeding lines to be used as parents or advanced in the breeding program for additional testing and finally variety release. Lines selected as parents must be genetically diverse and possess high mean performance for the desired traits. Breeders select superior lines from the base population to advance in testing toward the release of a variety. These lines must have a combination of beneficial alleles to warrant superior mean performance of desired traits across or within environments.

Traditional marker assisted selection was proposed as an artificial selection method to select individuals based on their allele constitution (Lande and Thompson, 1989). Marker assisted selection exploits the tight linkage between QTL and nearby markers and was suggested as an easier, faster, and more efficient selection method (Hospital, 2009). However, the use of traditional marker assisted selection has been limited in complex traits because of low power to detect QTL and bias in the estimated marker effects (Beavis, 1994; Melchinger et al., 1998; Bernardo, 2010). Another drawback of traditional marker assisted selection is that it splits the selection task into

two steps; identifying marker linked to QTL through QTL mapping followed by using linked markers to identify individuals carrying favorable alleles. The initial step of QTL mapping represents an impediment for the practical use of traditional marker assisted selection because of the need to assemble or create a mapping population, evaluate and genotype segregating progeny, and identify markers linked to QTL. Additional efforts are required to evaluate the mapped QTL to confirm they were not falsely declared (Bernardo, 2004, 2010; Navara and Smith, 2014). In addition, traditional methods of QTL mapping, such as bi-parental mapping are limited to the alleles from the two parents; therefore it does not represent the entire genetic diversity of the breeding program. Several studies have compared traditional marker assisted selection with phenotypic selection in several crop species including sweet corn (Yousef and Juvik, 2001), maize (Flint-Garcia et al., 2003; Abalo et al., 2009), wheat (Davies et al., 2006; Wilde et al., 2007), and soybeans (Lamkey at al., 2013). Mixed results were observed in these studies and in several cases phenotypic selection was superior to marker assisted selection (Flint Garcia et al., 2003; Davies et al., 2006; Wilde et al., 2007; Lamkey et al., 2013). A possible reason is that only significant markers are used in line selection, thus only a portion of the total genetic variance is captured by selected markers (Bernaro and Yu, 2007; Heffner et al., 2009).

Genomic selection (GS) is a relatively new practice in animal and plant breeding programs that does not require QTL mapping and aims to improve quantitative traits by exploiting abundant marker information (Meuwissen et al., 2001; Hayes et al., 2009; Lorenz et al., 2011). Genomic selection does not involve identifying significant markers

as is done in traditional marker assisted selection. In contrast, all markers are used, thereby reducing the two step process required for traditional marker assisted selection to a one step selection procedure that captures all the genetic variance explained by markers. Several genomic selection studies investigated various prediction models and found that ridge regression best linear unbiased prediction or RR-BLUP is equivalent to prediction models that manifest the underlying genetic architecture of a trait (Bayes Cπ and Bayes LASSO) or nonlinear prediction models that account for both additive and non-additive gene action (Gaussian and Exponential models) (Lorenz et al., 2012; Chapter 1).

Genomic selection enables a breeder to make selection decisions with higher accuracy compared to phenotypic or traditional marker assisted selection (Meuwissen et al., 2001; Bernardo and Yu, 2007; Lorenzana and Bernardo, 2009). With phenotypic selection, discrimination between breeding lines is performed at early stages of evaluation when phenotypic data is limited. Because genomic selection uses estimates of marker effects based on many breeding lines that were evaluated across multiple environments, it has the potential to provide more reliable breeding value estimates than when a limited number of trials is used in phenotypic selection. Genomic selection requires the availability of dense marker genotypes for the training and selection candidate populations and assembling a training population that has replicated phenotypes; which is a requirement of accurate estimates of marker effects.

Few studies have compared phenotypic selection (PS) and/or traditional marker assisted selection to genomic selection (GS) using simulated data (Bernardo and Yu, 2007; Jannink, 2010; Yabe et al. 2012) or empirical data (Asoro et al., 2013). In a

simulation study in maize, the response to genomic selection was found to exceed traditional marker assisted selection for low and moderate heritable traits (Bernardo and Yu, 2007). Additional studies using empirical data involving traits with different genetic architecture are needed to compare the effectiveness of genomic and phenotypic selection to identify superior progeny in breeding populations.

In this study, we compared GS and PS for disease resistance and yield in barley. Fusarium head blight or FHB, caused primarily by *Fusarium graminearum* Schwab, is one of the most serious diseases of barley in the Midwest USA. This disease causes marked reductions in both grain yield and quality—the latter occurring chiefly through the contamination of grain by the pathogen-produced-mycotoxin deoxynivalenol (DON) (Windels, 2000). Resistance to FHB and DON accumulation are quantitatively inherited and QTL associated with these traits have been identified on all barley chromosomes (de la Pena et al., 1999; Ma et al., 2000; Mesfin et al., 2003; Dahleen et al., 2003; Massman et al., 2011). Increasing yield is a major breeding objective in plant breeding. Yield in barley is a complex trait that is controlled by many genomic regions across chromosomes (Hayes et al., 1993; Zhu et al., 1999; Pauli et al., 2014). The main objective of this study was to compare the ability of GS and PS to identify the most superior individuals from several selection candidate populations. We compared selection response of GS and PS and assessed the relative efficiency GS over PS $RE_{GS:PS}$ using empirical data for yield, FHB resistance, and DON concentration. The effect of GS and PS on the rate of inbreeding was determined. The efficiency of indirect selection using GS and PS was investigated to improve a correlated trait.

Materials and Methods

Germplasm

To compare the effectiveness of GS and PS, we imposed selection on five sets of selection candidates. Each selection candidate set consisted of approximately 96 breeding lines that represent genetic diversity of the breeding program and that were entered into first year yield trials in the five years from 2006 to 2010. Prior to the first year yield trials, all breeding lines were advanced through single seed descent without selection to $F_{4:5}$. At this stage, the breeding lines were evaluated in FHB disease nurseries (two locations with two replicates per location as described below). Lines were selected to advance to first year yield trials based on FHB severity, DON concentration, heading date, and lodging if apparent. In a typical year, approximately 1,500 $F_{4:5}$ breeding lines were evaluated from which approximately 200 were advanced to first year yield trials. The 96 lines that comprised each of the selection candidate sets were a randomly selected subset of these lines. For FHB and DON we used all five selection candidate sets and for yield we used two selection candidate sets (2009 and 2010).

The training population used to generate predictions for GS was comprised of 168 breeding lines that were developed and evaluated prior to 2006 and included many of the parents that were used to develop the breeding lines in the selection candidate sets. This training population was described in detail in Chapter 1 and was referred to as the parent set. In that study the training population and selection candidate sets were used to evaluate the accuracy of genomic predictions.

The selected lines were the 10 best performing lines for yield, FHB severity, and DON concentration from each of the selection candidate sets. In each selection candidate set, the 10 best performing lines were selected based on phenotypic data or genomic predictions (Figure 1) as described below. The GS and PS selected lines for FHB and DON were reevaluated in FHB disease nurseries, and GS and PS selected lines for yield were evaluated in yield trials as described below. In some cases there was overlap between lines selected for FHB and DON or between lines selected by GS and PS. Regardless of whether a line was selected more than once, it was only represented once in a subsequent evaluation trial.

Phenotypic Evaluation of the Training Population, Selection Candidates, and Selected Lines

The number of breeding lines in all populations, time of evaluation, experimental design, and number of environments are displayed in (Table 1). The training population was evaluated in three locations in Minnesota where all lines were evaluated together in five trials for yield and three trials for FHB resistance and DON concentration (Chapter 1). Briefly, the lines in the training population were arranged in augmented incomplete block design experiments with two replications and four blocks per replication for yield, FHB resistance, DON concentration. Six common checks were included in each block to correct for spatial field variability. The six check cultivars (Drummond, Lacey, Quest, Rasmusson, Stellar, and Tradition) were randomly assigned to each block. In the yield trials, each line was represented once per block in two-row plots spaced 30 cm apart and

3 m in length. Yield was determined by harvesting individual plots with a Wintersteiger plot combine, weighing the grain, and expressing it as kg ha^{-1}. In the disease trials, each line was represented once per block in single row plots 1.8 m in length with 30 cm between rows. The plots were inoculated with the pathogen and mist irrigation was applied as described previously (Steffenson, 2003; Chapter 1). FHB severity was assessed 14 days after inoculation by estimating the percentage of diseased kernels on a random sample of 10 spikes per plot using the following assessment scale 0, 1, 3, 5, 10, 15, 25, 35, 50, 75, and 100%. DON concentration was determined on a 25g sample of the harvested grain from each plot by gas chromatography and mass spectrometry and expressed in parts per million according to the procedures of Mirocha et al. (1998). Correction for field spatial variability in the yield and FHB trials was carried out as described previously (Chapter 1).

The selection candidate sets were evaluated for yield, FHB resistance, and DON concentration between 2006 and 2010 in randomized complete block experiments with two replications in at least two locations in Minnesota and North Dakota (Table 1). Correction for spatial field variability and trial effects for parents and progeny sets were described previously (Chapter 1).

The GS and PS selected lines from each selection candidate set were revaluated in Minnesota and North Dakota in seven yield trials and four FHB disease trials in 2013. For yield, all lines were evaluated in augmented block design experiments with four blocks and three common checks (Quest, Tradition, and Lacey) repeated twice in each block. Lines and common checks were assigned randomly within each incomplete block.

For FHB resistance and DON concentration, the selected lines using the two selection schemes were evaluated together in four locations in Minnesota and North Dakota in randomized complete block design experiments with two replications. Four common checks (Quest, Robust, Stander, and MNBrite) were randomized twice in each replication.

Correction for spatial field variability in yield trials was done with the PROC MIXED procedure in SAS (9.3) using the three common checks as fixed effects and blocks as random effects in the mixed model equation (SAS Institute Inc., 2011; Wolfinger et al., 1997). Correction for spatial field variability in FHB disease trials was done using moving grid adjustment (R-package mvngGrAd, R development core team, 2013). A moving average window of 18 plants was used to determine the phenotypic performance of line in the center. For DON concentration, no spatial field correction was made as seed samples were combined among replications to obtain one DON measure for each line per trial.

For yield, the checks Tradition and Lacey and for FHB and DON, the checks Robust, Stander, and MNBrite were common between the selection candidates and selected trials (Rasmusson and Wilcoxson, 1983; Rasmusson et al., 1993; Horsley et al., 2006; Rasmusson et al., 2001). For all traits across selection candidates and selected trials, common checks were used to correct for variance in trial means using the MIXED procedure in SAS. In all experiments, genetic and error variance were calculated using the MIXED procedure in SAS. Broad sense heritability was estimated using the equation

$H^2 = \sigma_g^2/(\sigma_g^2 + \sigma_e^2/n)$; where σ_g^2 is genetic variance, σ_e^2 is the pooled error variance that includes GxE and residuals, and *n* is the number of trials.

Genotyping and Genomic Prediction

DNA genotyping for the training population and the selection candidates was performed in the USDA-ARS genotyping center in Fargo, ND with 1,536 SNPs referred to as BOPA1 (Close et al., 2009). The training population was screened to exclude SNP markers with minor allele frequency (MAF) < 0.01 and missing data > 10%. After screening markers with minor allele frequencies and missing data in the 168 training population, 984 markers were used construct a prediction model to predict the phenotypic performance of the selection candidate populations. The training population was used to predict the phenotypic performance of two sets of selection candidates for yield (2009 and 2010) and five sets of selection candidates for FHB severity and DON concentration (2006-2010) using ridge regression best linear unbiased prediction (RR-BLUP) implemented in the R-package rrBLUP (R development core team, 2012; Endelman, 2011).

RR-BLUP can be modeled as

$$y = \mu + \sum_{\{j=1\}}^{K} Z_j \, a_j + e$$

where $\boldsymbol{y}$ is the vector of individual phenotypes, $\boldsymbol{\mu}$ is the population mean, *K* is the number of markers, $\boldsymbol{Z}$ is the incidence matrix that links marker *j* genotypes to individuals, $\boldsymbol{a}$ is the

effect of marker j. In RR-BLUP, all markers are distributed with the same variance $N(0, \sigma_a^2)$. The variance of this distribution was estimated on the basis of marker and phenotypic data using restricted maximum likelihood (REML). The predictive ability of the prediction model was estimated as the correlation coefficient between genomic estimated breeding values (GEBV) and observed phenotypic values. Prediction accuracy was assessed by dividing the predictive ability by the square root of the heritability of the selection candidate population.

Evaluation of Selection Methods and Calculating Reponses to Selection

For GS and PS, breeding lines were ranked from best to the lowest performance in to select best performing breeding lines from each selection candidate set for each trait in each year separately. The 10 best performing lines for yield (highest), FHB severity (lowest), and DON concentration (lowest) were selected using GEBV and phenotypic values. Comparisons of trait means among selection candidates, GS, and PS selected lines were conducted using the means of phenotypic adjusted data (described above) by analysis of variance in the GLM procedure in SAS. Mean separation tests were based on Tukey's honestly significant difference (HSD; $\alpha = 0.05$). For both selection schemes, response to selection was expressed as the difference between the mean performance of the selected lines in the reevaluation trials (2013) and the mean of the complete selection candidate set evaluated in prior trials (2006-2010). Relative efficiency of GS to PS ($RE_{GS:PS}$) was calculated as the ratio of the response to GS divided by the response to PS. To investigate the effect of selection scheme on the genetic similarity, the simple

matching coefficient was calculated between the selected individuals for each selection scheme in each year separately using the R-package synbreed (R development core team, 2012; Wimmer et al., 2012)

In practical terms, the primary trait of interest with respect to FHB breeding is DON concentration; however, evaluating FHB severity is less expensive than assaying for DON in grain samples. We were therefore interested in evaluating several indirect methods of selecting for low DON. In addition to direct phenotypic selection for DON (DON_PS), we also used phenotypic selection for FHB (FHB_PS), genomic selection for FHB (FHB_GS), and genomic selection for DON (DON_GS). Analysis of variance was used to assess changes in DON concentration between selection schemes and selection candidate base populations using GLM procedure in SAS. Mean separation tests were based on Tukey's honestly significant difference (HSD; $\alpha = 0.05$). Responses to selection for the previous four selection methods were compared.

Results

We found that GS and PS performed similarly across multiple traits (Figure 2). Lines selected using GS for each year had a yield increase of 0.7 and 1.5% compared to the selection candidate base populations in 2009 and 2010, respectively, with an average yield increase of 1.1%. PS resulted in an increase of 4.5 and 2.0% in yield compared to 2009 and 2010 selection candidate populations, respectively, with an average yield increase of 3.3%. Although, GS and PS increased yield compared to the selection candidate base populations, the increase of yield was not significant (Table 2). For FHB

resistance, GS resulted in reduction in disease severity from 19.6 in the base populations to 17.1% in the GS selected lines (Table 2). Using GS, disease severity was reduced by 26.6, 14.4, 7.5, 8.9, and 4.6% in the selected lines from the five selection candidate populations with an average disease severity reduction in the GS selected lines of 12.4%. PS resulted in reduction in disease severity from the selection candidate populations from 19.6 to 17.5% in the PS selected lines. Using PS, disease severity was reduced by 22.8, 8.6, 16.8, 4.5, and 0.6% in the selected lines from the selection candidate populations between 2006 and 2010, respectively; with an average disease severity reduction in the PS selected lines of 10.4%. No significant differences were observed between GS and PS breeding schemes for FHB severity (Table 2). For DON concentration, GS resulted in overall significant reduction from 29.1 to 22.3 ppm in the GS selected lines (Table 2). Using GS, DON concentration was reduced by 34.7, 25.4, 21.2, 3.5, and 25% in the selected lines from the selection candidate populations in the five selection candidate sets (2006 – 2010), respectively, with an average DON reduction of 22%. PS resulted in overall significant reduction in DON concentration from 29.1 to 23.5 ppm in the PS selected lines. Using PS, DON concentration was reduced by 30.5, 25.4, 12.8, 14.0, and 15.4% in the selected lines from the selection candidate populations between 2006 and 2010, respectively, with an average DON reduction of 19.6%. No significant differences were observed between the selection schemes for DON concentration (Table 2).

For FHB, GS resulted in a significant increase in the genetic similarity in GS selected lines compared to the selection candidates and PS selected lines (p = is *0.01* and

0.04, respectively) (Figure 3). For yield and DON concentration no significant changes in the genetic similarity were observed among selection candidates, GS, and PS select lines.

In terms of response to selection, both selection schemes increased grain yield and reduced FHB resistance and DON concentration compared to the base populations (Table 3). In most cases the relative efficiency of GS exceeds PS (Table 3). Prediction accuracies for GS ranged from 0.36 to 0.66 for yield, from 0.62 to 0.80 for FHB resistance, and from 0.32 to 0.99 for DON concentration. Heritability estimates were not consistent among traits and years and ranged from 0.29 to 0.44 for yield, 0.32 to 0.68 for FHB resistance, and 0.47 to 0.63 for DON concentration. These heritability estimates are in general agreement with previously published studies for yield, FHB resistance, and DON concentration (Boukerrou and Rasmusson, 1990; Ma et al., 2000; Mesfin et al., 2003).

With regard to indirect selection for DON, we observed significant reductions of DON concentration from the base populations using all four selection methods (Figure 4). The four selection methods resulted in average DON concentration reduction of 20, 15.8, 21.9, and 19.6% for FHB_GS, FHB_PS, DON_GS, and DON_PS, respectively, from the base populations. No significant differences were observed between the four selection schemes. Responses to selection using the four selection methods showed highest response to selection when using DON_GS followed by FHB_GS (Figure 5).

Discussion

When would Genotypic Selection be Preferred over Phenotypic Selection?

In our breeding experiments, we observed that overall, GS and PS were similar in response for yield, FHB and DON, but in some cases one was superior to the other. Our results also indicated that GS and PS resulted in equal levels in genetic similarity for yield and DON concentration. In ten out of twelve selection candidates, if the r_a was greater than *H*, then the $RE_{GS:PS}$ was greater than one (Table 3). This suggests that the relative value of GS over PS is a function of the prediction accuracy compared to the square root of the heritability of the phenotypic selection data. GS prediction accuracy is influenced by many factors including trait architecture, training population size, and genetic distance between training and testing sets. GS accuracy can be improved by using dense marker coverage to capture all genetic variation and by the choice of a training population that is genetically related to the selection candidate population (Habier et al., 2007; Hayes et al., 2009; Chapter 1). The effectiveness of PS is directly related to *H*. H^2 is affected by genetic variance, environmental variability, and trait architecture. For a given trait, H^2 estimates could be increased by increasing genetic variance or reducing error variances. This could be possible by phenotypically evaluating breeding lines in more homogenous environments by correct for spatial and environmental variances. Both GS and PS breeding schemes were effective in reducing FHB severity and DON accumulation significantly from the selection candidate populations. However; both breeding schemes did not improve yield over the selection candidate populations (yield increase was not significant). Yield is a more complex trait and controlled by a larger

number of loci with smaller effects compared to FHB resistance and DON accumulation (Chapter 1), which can reduce selection efficiency. Previous empirical and simulation studies compared GS and PS as breeding schemes. Asoro et al. (2013) compared the two breeding schemes in two cycles of selection and observed an increase of β-glucan content in oat using GS and PS breeding schemes from the base population. They detected a slight improvement of using GS ($p = 0.08$) over PS.

Studies using empirical data usually test the efficiency of GS and PS breeding schemes over the short-term. On the other hand, simulation studies can be used to examine the long-term responses of different breeding schemes. Our results agree with Yab et al. (2013) when they found that the response to GS and PS are equal for traits with low and moderate heritability estimates. Jannink (2010) simulated 24 selection cycles and found that GS without weighting for low frequency marker alleles resulted in higher initial responses to selection for low and medium heritable traits compared to PS. However, the long-term PS resulted in greater selection response than unweighted GS which plateaued at earlier selection cycles.

Indirect Selection

Oftentimes selection for a trait is difficult or expensive to measure and breeders desire to measure traits in a convenient and less expensive way to evaluate a large number of breeding lines. This is certainly the case for DON as it requires specialized disease nurseries, harvesting, cleaning, grinding of grain samples, and laboratory analysis

to obtain the phenotypic value. The use of FHB resistance to indirectly select for low DON is possible as the correlation between these two traits phenotypes across the selection candidate populations in this study is about 0.70. Evaluating breeding lines for FHB severity is less expensive and time consuming than evaluating lines for DON. Predicting FHB severity or DON concentration is more convenient than actually measuring either trait. Generating prediction models for FHB will be easier as phenotypic data for DON is not required. Therefore predicted FHB severity, FHB severity, and predicted DON represent three indirect methods for selecting for lower DON. When we compared these three indirect methods to directly selecting using phenotypic DON data, we found that all the methods produced the desirable response to selection and were similar (Figure 4). These results suggest that a breeder could generate a prediction model for FHB or DON and successfully use it to select for lower DON. The use of indirect phenotypic selection is a function of the genetic correlation between observed phenotypic values for the primary and secondary traits and the heritability of the primary and secondary trait. On the other hand, the use of indirect genomic selection is a function of the genetic correlation between predicted genotypic values of the secondary trait and observed phenotypic values of the primary trait, prediction accuracy of the secondary trait, and heritability of the primary trait.

Costs of GS vs PS and breeding cycle time

Strategic allocation of resources for selection is essential and can shape the future direction of breeding programs. PS requires extensive evaluations of large populations over several environments to identify superior individuals. Additional time is needed for complex traits such as yield because this requires seed increase to evaluate selection candidates on large plots over multiple locations. On the other hand genomic selection relies on selecting the best performing breeding lines in a population based on genome-wide marker information in early generations (i.e., F_2 or F_3). Costs for GS, are associated with genotyping and phenotyping training populations and genotyping selection candidates (Chapter 1). Yield phenotypic evaluation in barley costs about $60 per entry when measuring over three locations. FHB disease assessment requires $40 ($10/plot) per entry to evaluate lines in two locations with two replication in each location. DON requires an additional $20 per entry to evaluate lines in two locations ($10 per grain sample). GS on the other hand costs about 20$ per entry when genotyping lines with 384 SNP array. Costs of genotyping are decreasing with the use of new technologies such as genotyping by sequencing (GBS) (Poland et al., 2012). Assuming equal response with GS and PS, GS should be substantially less expensive. Endelman et al., (2014) found that selection using GS is more effective than PS as genotyping costs decrease. In addition, the reduction in length of the breeding cycle associated with implementing GS can result in greater gain from selection compared with the use of phenotypic selection in breeding programs (Heffner et al., 2009).

A major advantage of implementing GS in breeding programs is the reduction of time required to release a new cultivar. GS can reduce the breeding cycle time by shortening time required to create new lines if the phenotypic performance of early generations is predicted for desired traits. In addition to reduction of breeding cycle time, GS can be used to predict the phenotypic performance of breeding lines when direct phenotyping of traits such as FHB and DON is time consuming. Implementing GS in the barley breeding program at the University of Minnesota resulted in reduction of time to release a new cultivar by more than one year. Giving that GS and PS breeding schemes are similar for their genetic gain, implementing GS in breeding programs can result in high genetic gain with reduction of both time and costs.

Table 1. Sets of breeding lines in the training population, selection candidate populations, and selected lines using GS and PS selection schemes. Number of breeding lines in each set (N), time of evaluation, experimental design, and number of trials are included for yield, FHB resistance, and DON concentration.

Trait	Breeding lines	*N*	Time of Evaluation	Exp. Design	No. trials
Yield	Training population	168	2009-2011	Augmented	5
	Selection candidates 2009	96	2009	RCB	2
	Selection candidates 2010	95	2010	RCB	3
	GS and PS selected lines	35	2013	Augmented	7
FHB	Training population	168	2009-2011	Augmented	3
	Selection candidates 2006	96	2006	RCB	3
	Selection candidates 2007	96	2007	RCB	3
	Selection candidates 2008	96	2008	RCB	3
	Selection candidates 2009	96	2009	RCB	3
	Selection candidates 2010	95	2010	RCB	2
	GS and PS selected lines	90	2013	RCB	3
DON	Training population	168	2009-2011	Augmented	3
	Selection candidates 2006	96	2006	RCB	3
	Selection candidates 2007	96	2007	RCB	2
	Selection candidates 2008	96	2008	RCB	4
	Selection candidates 2009	96	2009	RCB	3
	Selection candidates 2010	95	2010	RCB	2
	GS and PS selected lines	84	2013	RCB	4

Augmented: Augmented incomplete block experiment
RCB: Randomized complete block design experiment.

Table 2. Average of breeding lines across the selection candidate populations, genomic selection (GS) sets, and phenotypic selection (PS) sets for yield, FHB resistance, and DON concentration.

Population	Yield (Kg h^{-1})	FHB resistance (%)	DON concentration (ppm)
Selection candidates	8224.8 (A)	19.6 (A)	29.1 (A)
GS	8316.6 (A)	17.1 (B)	22.3 (B)
PS	8495.5 (A)	17.5 (B)	23.5 (B)

Letters indicate significant differences using Tukey's honestly significant difference ($\alpha = 0.05$).

Table 3. Responses to genomic selection (R_{GS}) and phenotypic selection (R_{PS}), relative efficiency of GS over PS ($RE_{GS:PS}$), prediction accuracy (r_a), and the square root of the heritability (H) for yield, FHB resistance, and DON concentration.

Trait	Year	R_{GS}	R_{PS}	$RE_{GS:PS}$	r_a	H
Yield	2009	58.79	378.29	0.16	0.36	0.54
	2010	538.70	593.50	0.90	0.66	0.66
FHB	2006	-5.86	-5.03	1.17	0.74	0.73
	2007	-2.80	-1.67	1.68	0.62	0.68
	2008	-1.50	-3.38	0.45	0.65	0.82
	2009	-1.64	-0.83	1.96	0.77	0.69
	2010	-0.83	0.11	7.38	0.80	0.57
DON	2006	-12.39	-10.90	1.14	0.99	0.75
	2007	-7.78	-7.76	1.00	0.85	0.75
	2008	-5.59	-3.38	1.65	0.97	0.69
	2009	-0.85	-3.36	0.25	0.32	0.79
	2010	-7.16	-4.41	1.62	0.41	0.69

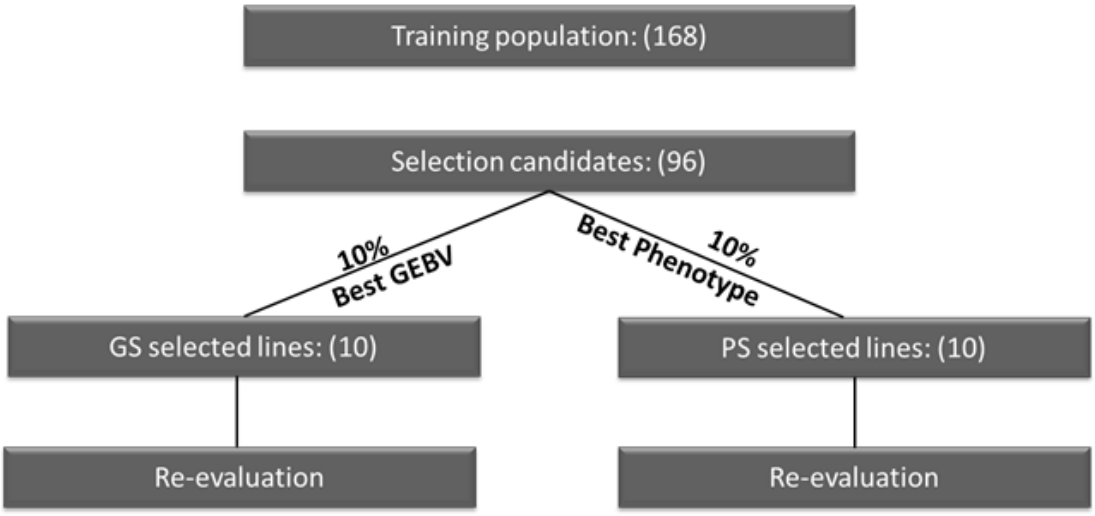

Figure 1. Breeding schemes for genomic selection (GS) and phenotypic selection (PS) for yield, Fusarium head blight (FHB) resistance, and Deoxynivalenol (DON) concentration. The training population (168 lines) was used to predict the performance of the selection candidates (96 lines each). 10% selection intensity was used to select the best performing individuals for GS and PS breeding schemes. The selected lines were re-evaluated again.

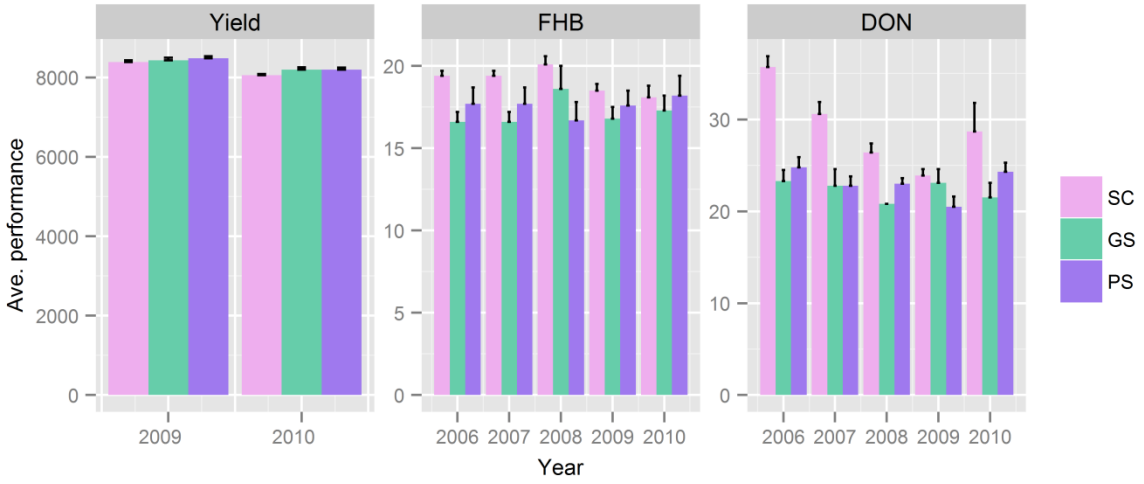

Figure 2. Average performance of yield (kg h^{-1}), FHB resistance (%), and DON concentration (ppm) in the selection candidates (SC), genomic selection (GS), and phenotypic selection (PS) sets.

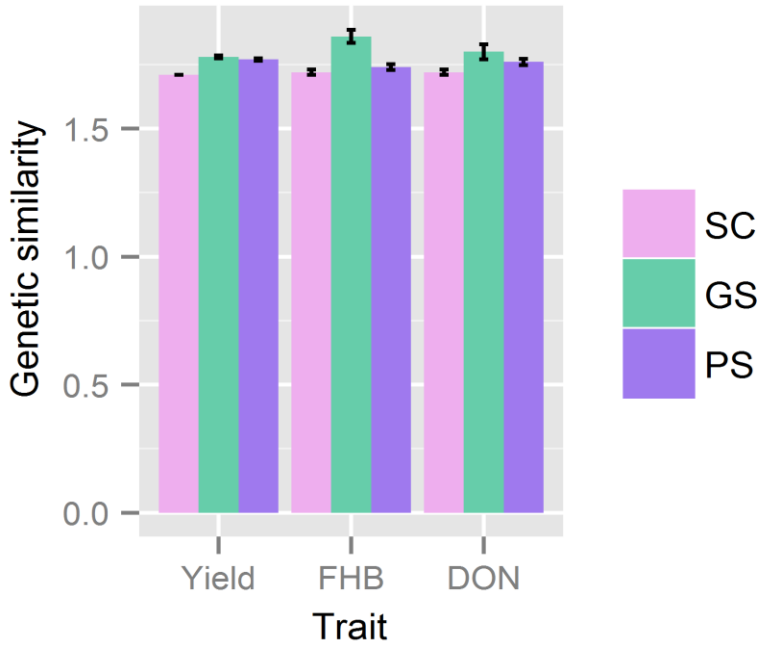

Figure 3. Average genetic similarity calculated as simple matching coefficient between lines in the selection candidates (SC), genomic selection (GS), and phenotypic selection (PS) sets.

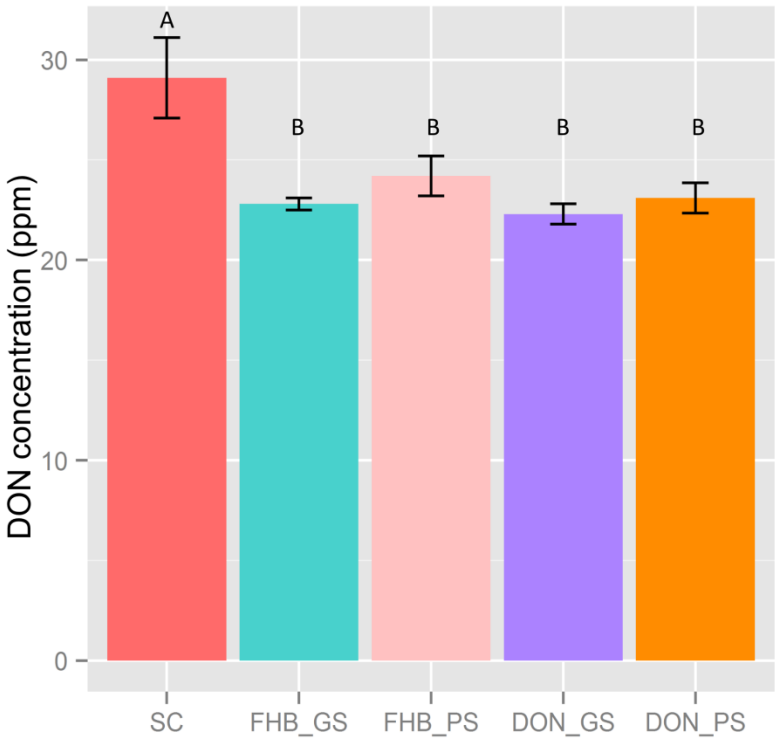

Figure 4. Mean DON concentration in (ppm) in the selection candidate populations (SC), lines selected using genomic selection for FHB (FHB_GS), phenotypic selection for FHB (FHB_PS), genomic selection for DON (DON_GS), and phenotypic selection for DON (DON_PS). Letters indicate significant differences based on Tukey's honestly significant difference ($\alpha = 0.05$).

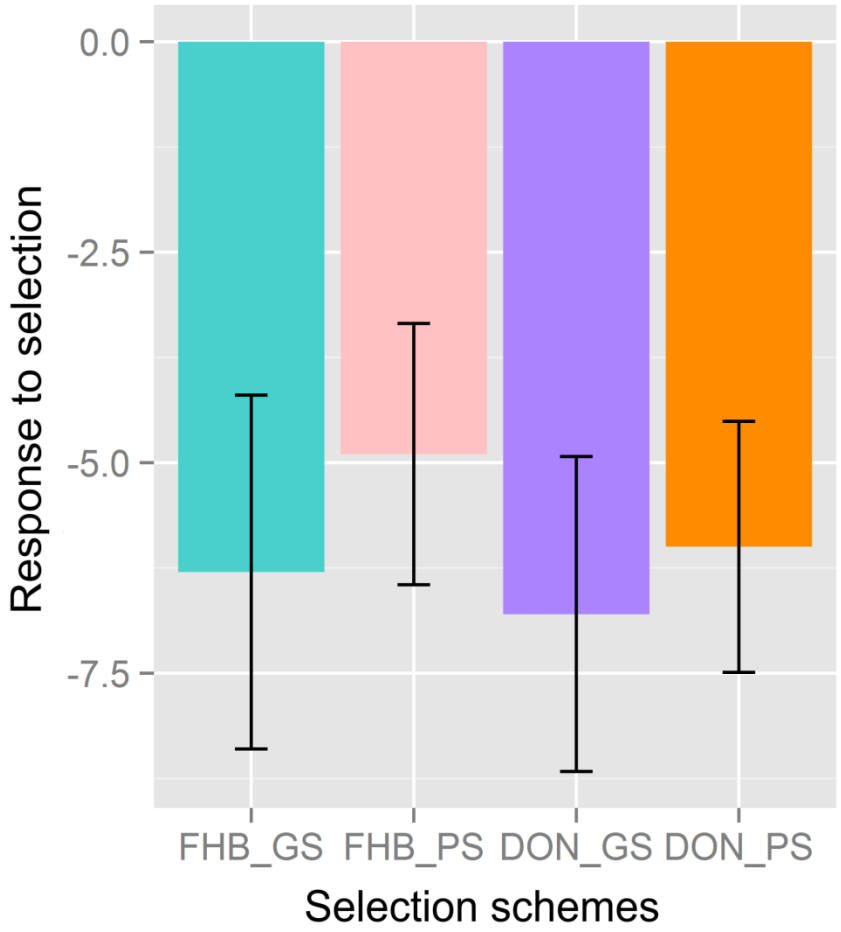

Figure 5. Response to selection when selecting lined using genomic selection for FHB (FHB_GS), phenotypic selection for FHB (FHB_PS), genomic selection for DON (DON_GS), and phenotypic selection for DON (DON_PS).

References

Abalo, G., P. Tongoona, J. Derera, and R. Edema. 2009. A comparative analysis of conventional and marker-assisted selection methods in breeding maize streak virus resistance in maize. Crop Sci. 49:509-520

Asoro, F.G., M.A. Newell, W.D. Beavis, M.P. Scott, and J.-L. Jannink. 2011. Accuracy and training population design for genomic selection on quantitative traits in elite North American oats. Plant Gen. 4:132–144. doi:10.3835/plantgenome2011.02.0007.

Asoro, G. Franco, M. Newell, W. Beavis, M.P. Scott, N. Tinker, and J.-L. Jannink. 2013. Genomic, Marker-Assisted, and Pedigree-BLUP Selection Methods for β-Glucan Concentration in Elite Oat. Crop Sci. 53:1894-1906.

Barrett, J.C., J. Maller, and M.J. Daly.2005. Haploview: analysis and visualization of LD and haplotype maps. Bioinformatics 21:263-265.

Barton, N.H., and S.P. Otto. 2005. Evolution of Recombination Due to Random Drift. Genetics 169:2353–2370. doi: 10.1534/genetics.104.032821.

Beavis, W.D. 1994. The power and deceit of QTL experiments: Lessons from comparative QTL studies. In: D. B. Wilkinson (ed.) 49th Ann Corn Sorghum Res Conf. Am Seed Trade Assoc., Chicago, IL. pp. 250-266.

Bernardo, R. 2004. What proportion of declared QTL in plants are false? Theor. Appl. Genet. 104:419-424.

Bernardo, R., and J. Yu. 2007. Prospects for genome-wide selection for quantitative traits in maize. Crop Sci. 47:1082–1090.

Bernardo, R. 2008. Molecular markers and selection for complex traits in plants: Learning from the last 20 years. Crop Sci. 48:1649-1664. doi:10.2135/cropsci2008.03.0131.

Bernardo, R. 2010. Breeding for Quantitative Traits in Plants. Woodbury, MN: Stemma Press.

Blake, V.C., J.G. Kling, P.M. Hayes, and J. L. Jannink. 2012 The Hordeum Toolbox -The Barley Coordinated Agricultural Project genotype and phenotype resource. The Plant Genome 5:81-91.

Boukerrou, L., and D. Rasmusson. 1990. Breeding for high biomass yield in spring barley. Crop Sci. 30:31-35.

Castro, A.J., F. Capettini, A.E. Corey, T. Filichkina, P.M. Hayes, A. Kleinhofs, D. Kudrna, K. Richardson, S. Sandoval-Islas, C. Rossi, and H. Vivar. 2003. Mapping and pyramiding of qualitative and quantitative resistance to stripe rust in barley. Theor. Appl. Genet. 107:922-930.

Chen, C.Y., I. Misztal, I. Aguilar, S. Tsuruta, T. H. E. Meuwissen, S. E. Aggrey, T. Wing, and W. M. Muir. 2012. Genome-wide marker-assisted selection combining all pedigree phenotypic information with genotypic data in one step: An example using broiler chickens. J. Anim. Sci. 89:23-28.

Close, T.J., P.R. Bhat, S. Lonardi, Y. Wu, N. Rostoks, L. Ramsay, A. Druka, N. Stein, J.T. Svensson, S. Wanamaker, S. Bozdag, M.L. Roose, M.J. Moscou, S. Chao, R.K. Varshney, P. Szuecs, K. Sato, P.M. Hayes, D.E. Matthews, A. Kleinhofs, G.J. Muehlbauer, J. DeYoung, D.F. Marshall, K. Madishetty, R.D. Fenton, P. Condamine, A.

Graner, and R. Waugh. 2009. Development and implementation of high-throughput SNP genotyping in barley. BMC Genomics 10:582.

Collins, H.M., J.F. Panozzo, S.J. Logue, S.P. Jefferies, and A.R. Barr. 2003. Mapping and validation of chromosome regions associated with high malt extract in barley (Hordeum vulgare L.). Aust J Agric Res. 54:1223-1240.

Combs, E., and R. Bernardo. 2013. Accuracy of genomewide selection for different traits with constant population size, heritability, and number of markers. Plant Gen. 6:1-7. doi: 10.3835/plantgenome2012.11.0030.

Condón, F., C. Gustus, D.C. Rasmusson, and K.P. Smith. 2008. Effect of Advanced Cycle Breeding on Genetic Diversity in Barley Breeding Germplasm. Crop Sci. 48:1027-1036.

Condon, F., D.C. Rasmusson, E. Schiefelbein G. Velasquez, and K.P. Smith. 2009. Effect of Advanced Cycle Breeding on Genetic Gain and Phenotypic Diversity in Barley Breeding Germplasm. Crop Sci. 49: 1751-1761.

Crossa, J., G. de los Campos, P. Perez, D. Gianola, J. Burgueno, J. Luis Araus, D. Makumbi, R.P. Singh, S. Dreisigacker, J. Yan, V. Arief, M. Banziger, and H. Braun. 2010. Prediction of genetic values of quantitative traits in plant breeding using pedigree and molecular markers. Genetics 186:713-724.

Daetwyler, H.D., R. Pong-Wong, B. Villanueva, and J.A. Woolliams. 2010. The impact of genetic architecture on genome-wide evaluation methods. Genetics 185:1021–1031. doi:10.1534/genetics.110.116855.

Dahleen, L.S., H.A. Agrama, R.D. Horsley, B.J. Steffenson, P.B. Schwarz, A. Mesfin, and J.D. Franckowiak. 2003. Identification of QTLs associated with Fusarium head blight resistance in Zhedar 2 barley. Theor. Appl. Genet. 108:95-104

Davies, J., W.A. Berzonsky, G.D. Leach. 2006. A comparison of marker-assisted selection and phenotypic selection for high grain protein content in spring wheat. Euphytica 152: 117-134

de la Pena, R.C., K.P. Smith, F. Capettini, G.J. Muehlbauer, M. Gallo- Meagher, R. Dill-Macky, D.A. Somers, and D.C. Rasmusson. 1999. Quantitative trait loci associated with resistance to Fusarium head blight and kernel discoloration in barley. Theor. Appl. Genet. 99: 561–569.

de los Campos, G., D. Gianola, and G.J.M. Rosa. 2009. Reproducing kernel Hilbert spaces regression: A general framework for genetic evaluation. J. Anim. Sci. 87:1883–1887. doi:10.2527/jas.2008-125.

de Roos, a.P.W., B.J. Hayes, and M.E. Goddard. 2009. Reliability of genomic predictions across multiple populations. Genetics 183:1545-1553.

de Roos, a.P.W., B.J. Hayes, R.J. Spelman, and M.E. Goddard. 2008. Linkage Disequilibrium and Persistence of Phase in Holstein–Friesian, Jersey and Angus Cattle. Genetics 183:1545-1553. Genetics 179:1503–1512. doi: 10.1534/genetics.107.084301.

Dekkers, J.C.M. 2004. Commercial application of marker- and gene-assisted selection in livestock: strategies and lessons. J. Anim. Sci. 82: E313-E328.

Endelman, J.B. 2011. Ridge regression and other kernels for genomic selection with R package rrBLUP. Plant Genome 4:250–255. doi:10.3835/plantgenome2011.08.0024.

Endelman, J.B.,G.A. Atlin, Y. Beyene, K. Semagn, X. Zhang, M. Sorrells, and J.-L. Jannink. 2014. Optimal design of preliminary yield trials with genome-wide markers. Crop Sci. 54:48-59.

Falconer, D.S., and T.F.C. Mackay. 1996. Introduction to quantitative genetics. 4th ed. Longman Technical and Scientific, Essex, UK.

Fang, Z., A. Eule-Nashoba, C. Powers, T.Y. Kono, S. Takuno, P.L. Morrell, and K.P. Smith. 2013. Comparative Analyses Identify the Contributions of Exotic Donors to Disease Resistance in a Barley Experimental Population. G3 3: 1945–1953. doi: 10.1534/g3.113.007294

Flint-Garcia, S.A., L.L. Darrah, M.D. McMullen, B.E. Hibbard. 2003. Phenotypic versus marker-assisted selection for stalk strength and second generation European corn borer resistance in maize. Theor. Appl. Genet. 107:1331-1336.

Gianola, D., and J. B. C. H.M. van Kaam. 2008. Reproducing kernel Hilbert spaces regression methods for genomic assisted prediction of quantitative traits. Genetics 178: 2289–2303.

Gianola, D. 2013. Priors in whole-genome regression: The Bayesian alphabet returns. Genetic 194:573-596.

Goddard, M.E., and B.J. Hayes. 2007. Genomic selection. J. Anim. Breed. Genet. 124:323–330. doi:10.1111/j.1439-0388.2007.00702.x.

Habier, D., R.L. Fernando, and J.C.M. Dekkers. 2007. The impact of genetic relationship information on genome-assisted breeding values. Genetics 177:2389-2397.

Hayes, B.J., P.J. Bowman, A.C. Chamberlain, and M.E. Goddard. 2009. Genomic selection in dairy cattle: Progress and challenges. J. Dairy Sci. 92:433-443. doi:10.3168/jds.2008-1646.

Hayes, B.J., J. Pryce, A.J. Chamberlain, P.J. Bowman, and M.E. Goddard. 2010. Genetic architecture of complex traits and accuracy of genomic prediction: coat colour, milk-fat percentage and type in holstein cattle as contrasting model traits. PLoS Genet 6: e1001139. doi: 10.1371/journal.pgen.1001139.

Hayes, B.J., P.J. Bowman, A.C. Chamberlain, and M.E. Goddard. 2009. Genomic selection in dairy cattle: Progress and challenges. J. Dairy Sci. 92:433-443. doi:10.3168/jds.2008-1646.

Hayes, P.M., B.H. Liu, S.J. Knapp, F. Chen, B. Jones, T. Blake, J. Franckowiak, D. Rasmusson, M. Sorrells, S.E. Ullrich, D. Wesenberg, and A. Kleinhofs. 1993. Quantitative trait locus effects and environmental interaction in a sample of North American barley germplasm. Theor. Appl. Genet. 87:392-401.

Heffner, E.L., M.E. Sorrells, and J. L-Jannink. 2009. Genomic selection for crop improvement. Crop Sci. 49:1-12. doi: 10.2135/cropsci2008.08.0512.

Heffner, E.L., A.J. Lorenz, J. Jannink, and M.E. Sorrells. 2010. Plant breeding with genomic selection: Gain per unit time and cost. Crop Sci. 50:1681-1690. doi:10.2135/cropsci2009.11.0662.

Heffner, E.L., J.-L. Jannink, and M.E. Sorrells. 2011. Genomic selection accuracy using multifamily prediction models in a wheat breeding program. Plant Gen. 4:65–75. doi:10.3835/plantgenome.2010.12.0029.

Hofheinz, N., D. Borchardt, K. Weissleder, and M. Frisch. 2012. Genome-based prediction of test cross performance in two subsequent breeding cycles. Theor. Appl. Genet. 125:1639–1645.

Horsley, R.D., J.D. Franckowiak, P.B. Schwarz, and B.J. Steffenson. Registration of 'Drummond' barley. Crop Sci. 2002. 42:664–665.

Horsley, R.D., D. Schmierer, C. Maier, D. Kudrna, C.A. Urrea, B.J. Steffenson, P.B. Schwarz, J.D. Franckowiak, M.J. Green, B. Zhang, and A. Kleinhofs. 2006. Identification of QTLs associated with Fusarium head blight resistance in barley accession CIho 4196. Crop Sci. 46:145-156.

Horsley, R.D., R.D., J.D. Franckowiak, P.B. Schwarz, S.M. Neate. 2006. Registration of 'Stellar-ND' Barley. Crop Sci. 46:980–981.

Hospital, F. 2009. Challenges for effective marker-assisted selection in plants. Genetica 136:303–310.

Jannink, J.-L. 2010. Dynamics of long-term genomic selection. Genet. Sel. Evol. 42:35. doi:10.1186/1297-9686-42-35

Jannink, J.-L., A. J. Lorenz, and H. Iwata. 2010. Genomic selection in plant breeding: from theory to practice. Brief. Funct. Genomics. 9(2): 166–177.

Kang, H.M., N.A. Zaitlen, C.M. Wade, A. Kirby, D. Heckerman, M.J. Daly, and E. Eskin. 2008. Efficient control of population structure in model organism association mapping. Genetics 178:1709–1723. doi:10.1534/genetics.107.080101.

Kizilkaya, K., R.L. Fernando, and D.J. Garrick. 2010. Genomic prediction of simulated multibreed and purebred performance using observed fifty thousand single nucleotide polymorphism genotypes. J. Anim. Sci. 88:544-551.

Lamkey, C.M., T.C. Helms, and R.J. Goos. 2013. Marker-assisted versus phenotypic selection for iron-deficiency chlorosis in soybean. Euphytica 194:67-78.

Lande, R., and R. Thompson. 1990. Efficiency of marker-assisted selection in the improvement of quantitative traits. Genetics 124: 743-756.

Legarra, A., C. Robert-Granie, E. Manfredi, and J. M. Elsen. 2008. Performance of genomic selection in mice. Genetics 180:611-618.

Liu, P., J. Zhu, X. Lou, and Y. Lu. 2003. A method for marker-assisted selection based on QTLs with epistatic effects. Genetica 119:75–86.

Lorenz, A.J., S. Chao, F.G. Asoro, E.L. Heffner, T. Hayashi, H. Iwata, K.P. Smith, M.E. Sorrells, and J.-L. Jannink. 2011. Genomic selection in plant breeding: Knowledge and prospects. Adv. Agron. 110:77–123. doi:10.1016/B978-0-12-385531-2.00002-5.

Lorenz, A.J., K.P. Smith, and J.-L. Jannink. 2012. Potential and optimization of genomic selection for Fusarium head blight resistance in six-row barley. Crop Sci. 52:1609-1621. doi 10.2135/cropsci2011.09.0503.

Lorenz, A. J. 2013. Resource allocation for maximizing prediction accuracy and genetic gain of genomic selection in plant breeding: A simulation experiment. G3. 3:481-491. doi: 10.1534/g3.112.004911.

Lorenzana, R.E., and R. Bernardo. 2009. Accuracy of genotypic value predictions for marker-based selection in biparental plant populations. Theor. Appl. Genet. 120:151–161. doi:10.1007/s00122-009-1166-3.

Luan, T., J.A. Woolliams, S. Lien, M. Kent, M. Svendsen, and T.H. Meuwissen. 2009. The accuracy of genomic selection in Norwegian red cattle assessed by cross-validation. Genetics 183:1119-1126. doi: 10.1534/genetics.109.107391.

Ma, Z., B.J. Steffenson, L.K. Prom, and N.L.V. Lapitan. 2000. Mapping quantitative trait loci for Fusarium head blight resistance in barley. Phytopathology 90:1079-1088. doi: 10.1094/PHYTO.2000.90.10.1079.

Massman, J., B. Cooper, R. Horsley, S. Neate, R. Dill-Macky, S. Chao, Y. Dong, P. Schwarz, G.J. Muehlbauer and K.P. Smith. 2011. Genome-wide association mapping of Fusarium head blight resistance in contemporary barley breeding germplasm. Mol. Breed. 27:439-454.

Melchinger, A.E., H.F. Utz, and C.C. Schon. 1998. Quantitative trait locus (QTL) mapping using different testers and independent population samples in maize reveals low power of QTL detection and large bias in estimates of QTL effects. Genetics 149:383–403

Mesfin, A., K.P. Smith, R. Waugh, R. Dill-Macky, C.K. Evans, C.D. Gustus, and G.J. Muehlbauer. 2003. Quantitative trait loci for Fusarium head blight resistance in barley detected in a two-rowed by six-rowed population. Crop Sci. 43: 307-318. doi:10.2135/cropsci2003.3070.

Meuwissen, T.H.E., B.J. Hayes, and M.E. Goddard. 2001. Prediction of total genetic value using genome-wide dense marker maps. Genetics 157:1819–1829.

Mirocha, C.J., E. Kolaczkowski, W. Xie, H. Yu, and H. Jelen. 1998. Analysis of deoxynivalenol and its derivatives (batch and single kernel) using gas chromatography/mass spectrometry. Journal of Agricultural and Food Chemistry. 46:1414-1418.

Nei, M. 1987. Molecular Euolutionary Genetics. Columbia University Press, New York.

Pauli, D., G.J. Muehlbauer, K. Smith, B. Cooper, Obert D.E., S.E. Ullrich, and T.K. Blak. 2014. Association mapping of agronomic QTLs in U.S. spring barley breeding germplasm. Plant Gen. 7:-. doi:10.3835/plantgenome2013.11.0037

Pfaffelhuber, P., A. Lehnert, and W. Stephan. 2008. Linkage disequilibrium under genetic hitchhiking in finite populations. Genetics 179:527–537.

Piepho, H.P., 2009. Ridge regression and extensions for genomewide selection in maize. Crop Sci. 49:1165–1176. doi:10.2135/cropsci2008.10.059.

Poland, J., J. Endelman, J. Dawson, J. Rutkoski, S. Wu, Y. Manes, S. Dreisigacker, J. Crossa, H. Sánchez-Villeda, M. Sorells, and J.-L. Jannink. 2012. Genomic selection in wheat breeding using genotyping-by-sequencing. The Plant Genome 5:103-113. doi:10.3835/plantgenome2012.06.0006

Pritchard, J.K., M. Stephens, N.A. Rosenberg, and P. Donnelly. 2000. Association mapping in structured populations. Am. J. Hum. Genet. 67:170–181.

R Development Core Team. 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.r-project.org.

Rasmusson, D. C., K. P. Smith, R. Dill-Macky, E. L. Schiefelbein, and J. V. Wiersma . 2001. Registration of 'Lacey' Barley. Crop Sci 41:1991.

Rasmusson, D.C., R.D. Wilcoxson, and J.V. Wiersma. 1993. Registration of 'Stander' barley. Crop Sci. 33:1403.

Rasmusson, D.C., and R.D. Wilcoxson. Registration of 'Robust' barley. 1983. Registration of 'Robust' barley. Crop Sci. 23:1216

Rutkoski, J., J. Benson, Y. Jia, G. Brown-Guedira, J.-L. Jannink and M. Sorrells. 2012. Evaluation of genomic prediction methods for Fusarium head blight resistance in wheat. Plant Gen. 5:51-61.

SAS Institute. 2011. The SAS system for Windows. Version 9.3. SAS Inst., Cary, NC.

Slotta, T.A., L. Brady, S. Chao. 2008. High throughput tissue preparation for large-scale genotyping experiments. Mol. Ecol. Resour. 8(1):83-87. doi: 10.1111/j.1471-8286.2007.01907.x.

Smith, K.P., A. Budde, R. Dill-Macky, D.C. Rasmusson, E. Schiefelbein, B. Steffenson, J.J. Wiersmaa, J.V. Wiersmad, and B. Zhang. 2013. Registration of 'Quest' spring malting barley with improved resistance to Fusarium head blight. J. Plant Reg. 7:125-129. doi:10.3198/jpr2012.03.0200crc.

Smith, K.P., D.C. Rasmusson, E. Schiefelbein, J.J. Wiersma, J. V Wiersma,. A. Budde, R. Dill-Macky, B.Steffenson, 2010. Registration of 'Rasmusson' Barley 2010. J. Plant Reg. 4:164-167.

Steffenson, B.J. (2003). Fusarium head blight of barley: impact, epidemics, management, and strategies for identifying and utilizing genetic resistance. Fusarium head blight of wheat and barley: 241-295.

Technow, F., A. Bürger, A. E. Melchinger. 2013. Genomic prediction of northern corn leaf blight resistance in maize with combined or separated training sets for heterotic groups. G3 3:197-203. doi: 10.1534/g3.112.004630.

Toosi, A., R. Fernando and J. Dekkers. 2010. Genomic selection in admixed and crossbred populations. J. Anim. Sci. 88: 32-46.

Wang, D., S.I. El-Basyoni, S.P. Baenziger, J. Crossa, K.M. Eskridge, I. Dweikat. 2012. Prediction of genetic values of quantitative traits with epistatic effects in plant breeding populations. Heredity 109:313–319. doi:10.1038/hdy.2012.44.

Weir, B.S., C.C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. Evolution 38:1358-1370.

Wilde, F., V. Korzun, E. Embeyer, H.H. Geiger, and T. Miedaner. 2007. Comparison of phenotypic and marker-based selection of Fusarium head blight resistance and DON contents in spring wheat. Mol. Breed. 19:375-370.

Wimmer, V., T. Albrecht, H.-J. Auinger, and C.-C. Schön. 2012. synbreed: a framework for the analysis of genomic prediction data using R. Bioinformatics 28:2086-2087. doi: 10.1093/bioinformatics/bts335.

Wimmer, V., C. Lehermeier, T. Albrecht, H.-J. Auinger, Y. Wang, and C.-C. Schön. 2013. Genome-Wide Prediction of Traits with Different Genetic Architecture Through Efficient Variable Selection. Genetics 195:573-587. doi: 10.1534/genetics.113.150078.

Windels, C.E. 2000. Economic and social impacts of Fusarium head blight: Changing farms and rural communities in the northern great plains. Phytopathology 90:17–21.

Windhausen, V. S., G.A. Atlin, J.M. Hickey, J. Crossa, J.-L. Jannink, M.E. Sorrells, B. Raman, J.E. Cairnst, A. Tarekegne, K. Semagen, Y. Beyene, P. Grudloyma, F. Technow, C. Riedelsheimer, and A. Melchinger. 2012. Effectiveness of Genomic Prediction of Maize Hybrid Performance in Different Breeding Populations and Environments. G3 2:1427–1436. doi:10.1534/g3.112.003699.

Wolfinger, R.D., W.T. Federer, and O. Cordero-Brana. 1997. Recovering information in augmented designs, using SAS PROC GLM and PROC MIXED. Agron. J. 89:856-859

Xu, Y. and J.H. Crouch. 2008. Marker-Assisted Selection in Plant Breeding: From Publications to Practice. Crop Sci. 48:391-407. doi:10.2135/cropsci2007.04.0191.

Yabe, S., R. Ohsawab, and H. Iwata. 2013. Potential of genomic selection for mass selection breeding in annual allogamous crops. Crop Sci. 53:95–105.

Yousef, G.G., and J.A. Juvik. 2001. Comparisons of phenotypic and marker-assisted selection for quantitative traits in sweet corn. Crop Sci. 41:645-655

Zhao, K., C.-W. Tung, G.C. Eizenga, M.H. Wright, M.L. Ali, A.H. Price, G. Norton, M.R. Islam, A. Reynolds, J. Mezey, A.M. MsClung, C.D. Bustamante, and S. McCouch. 2011. Genome-wide association mapping reveals a rich genetic architecture of complex traits in Oryza sativa. Nature comms. 2:467. doi:10.1038/ncomms1467.

Zhao, Y., M. Gowda, W. Liu, T. Würschum, H.P. Maurer, F.H. Longin, N. Ranc, and J.C. Reif. 2012. Accuracy of genomic selection in European maize elite breeding populations. Theor. Appl. Genet. 124:769-776.

Zhong, S., J.C.M. Dekker, R.L. Fernando, and J.-L. Jannink. 2009. Factors affecting accuracy from genomic selection in populations derived from multiple inbred lines: a barley case study. Genetics 182: 355–364.

Zhu, H., G. Briceño, R. Dovel, P.M. Hayes, B.H. Liu, C.T. Liu , T. Toojinda, and S.E. Ullrich. 1999. Molecular breeding for grain yield in barley: an evaluation of QTL effects in a spring barley cross. Theor. Appl. Genet. 98:772-779.

Appendix

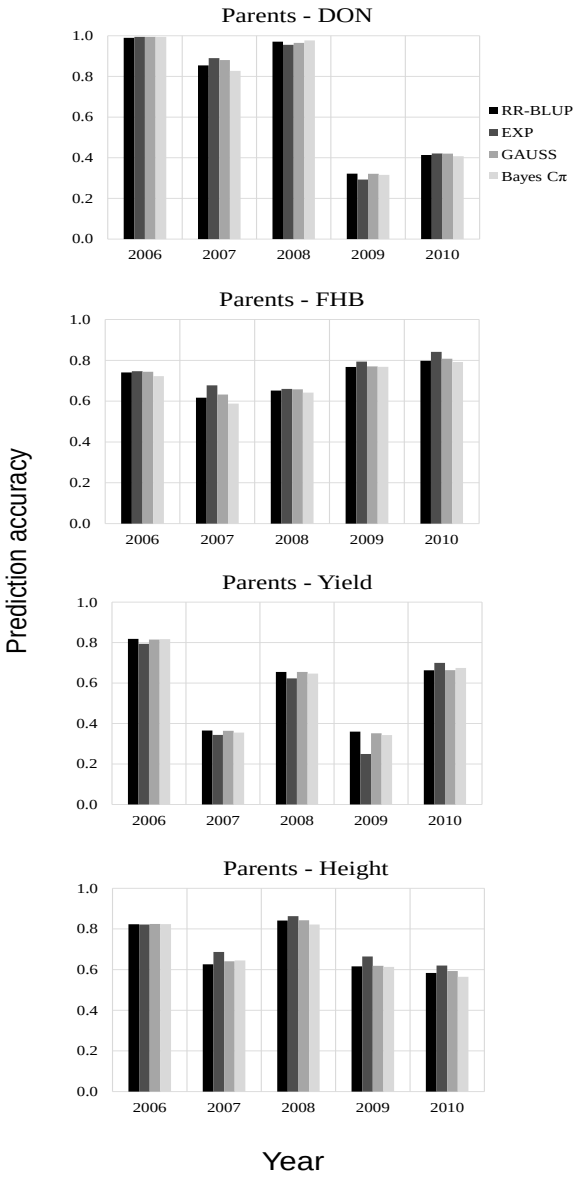

Figure S1. Prediction accuracy for A) DON accumulation, B) FHB resistance, C) yield, and D) plant height using RR-BLUP, Exponential kernel method, Gaussian kernel method, and Bayes Cπ when using the parent set as a training population to predict the five progeny sets.

Table S1. Number of experimental trials for the parent and five progeny sets for deoxynivalenol (DON) accumulation, Fusarium head blight (FHB) resistance, yield, and plant height. Each line was replicated twice in each experiment.

	Number of trials			
Population	**DON**	**FHB**	**Yield**	**Plant height**
Contemporary parent data	3	3	5	3
2006	3	3	3	3
2007	2	3	3	3
2008	4	3	2	3
2009	3	3	2	3
2010	2	2	3	2

DON, Deoxynivalenol accumulation
FHB, Fusarium head blight resistance