Association Mapping of Hagberg Falling Number in Hard White Spring Wheat Accessions

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Introduction

➢The Hagberg falling number (FN) (Hagberg, 1960) has been used as a criterion of end-use quality of wheat (Belitz et al. 2004); and wheat grains with low FN (< 300 sec.) tend to have poor end-use quality, such as stickiness. Grain with low FN is either rejected or the price discounted.

➢ Pre-harvest sprouting (PHS) and late maturity α-amylase (LMA) induce low FN in wheat grains, and can be differentiated with a α-amylase activity test using a half kernel method (Mares and Mrva, 2008).

➢ FN testing is expensive, time-consuming, and significantly affected by environments and growing conditions. Therefore, molecular markers associated with FN can be used to accelerate removal of low FN lines in the breeding process.

➢ Few studies have been done to identify molecular markers associated with FN although some of studies have identified QTL and markers associated with PHS or LMA.

➢ FN screening of hard white spring (HWS) wheat accessions in National Small Grain Collection (NSGC) has not been done previously and these accessions are genetically diverse and thus ideal for identifying molecular markers associated with FN using an association mapping approach.

Objectives

➢ Evaluate genetic variation for FN in 123 HWS wheat accessions

➢ Discern the causes of low FN: either PHS or LMA or both

➢ Identify FN-associated SNP markers by association mapping

➢ Identify novel accessions resistant to PHS and/or LMA

Materials and Methods

➢123 HWS accessions were used from NSGC.

➢ Accessions were grown under three field treatments in Aberdeen, ID in 2011: normal irrigation and normal nitrogen fertilizer (T1) ; limited irrigation and normal nitrogen (T2); limited irrigation and low nitrogen (T3).

➢ FN was tested using a Perten Falling Number 1700 instrument.

➢ Correlation and ANOVA analyses were conducted using R 2.15.1 (R Core Team 2012).

➢ Half kernel α-amylase (AA) was tested using the Megazyme Ceralpha kit only for those accessions with FN less than 300 sec.

➢ Principal component analysis (PCA) was used to explore the structure of the population.

➢ Association mapping of FN was conducted using Tassel 3.0 (Bradbury et al. 2007).

➢ General linear model (GLM) was used with the Q matrix from PCA analysis; the Bonferroni correction (α = 0.1) was used to detect significant markers.

Results and Discussion (1)

➢ The FN variation of all accessions showed a normal distribution under all three treatments, indicating it is a quantitative genetic trait (Fig 1).

➢ More accessions with FN less than 300 seconds were identified in T1 (28) than in T2 (10) and T3 (12) (Table 1).

➢ Multiple comparisons showed that FN mean of T1 was significantly lower than that of T2 and T3, while FN for T2 and T3 were not significantly different (Table 1). This result suggests that irrigation decreased the FN, possibly due to increased lodging.

➢ The correlation of FN between T2 vs. T3 was greater (*r* = 0.80) than the correlation between T1 vs. T2, and T1 vs. T3 (Fig 1).

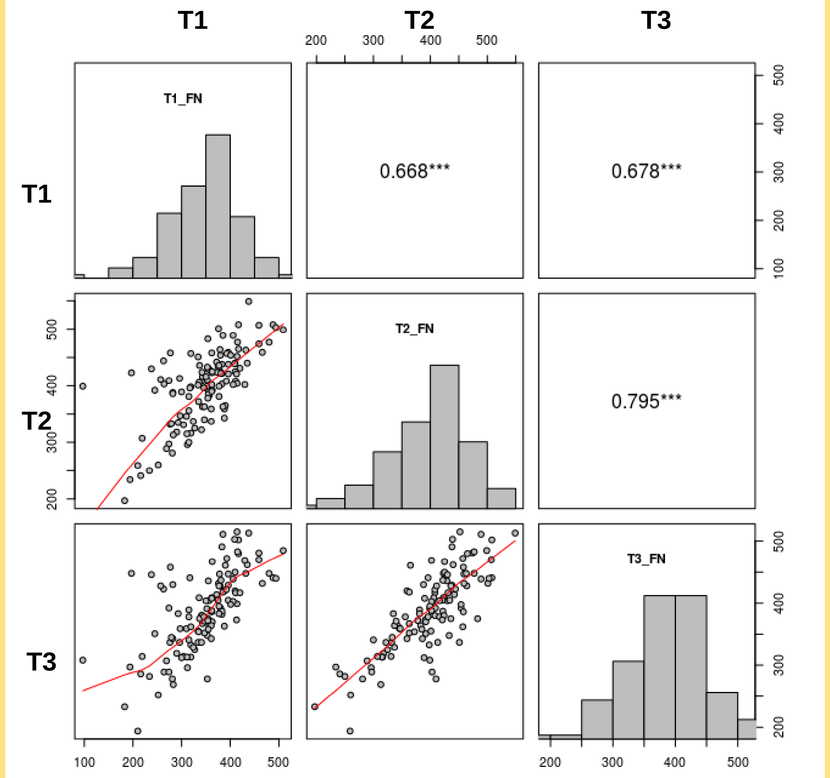

Fig 1. Histograms (the diagonal), correlation coefficients (the upper diagonal, *** significant at α = 0.001) and scatter plot and loess smoothing line (the lower diagonal) of the falling number (FN) across three treatments (T1, T2, T3)

Table 1. Mean and range of FN (sec.) of 123 HWS accessions grown under three treatments in Aberdeen, ID in 2011

Treatment	FN			No. of accessions with FN <300
	Mean	Min	Max	
T1	347b	97	509	28
T2	398a	197	549	10
T3	388a	194	515	12

Table 2. α-amylase activity ratio (AAR, head/tail) of 30 accessions with FN less than 300 sec. grown under three treatments in Aberdeen, ID in 2011

Treatment	AAR		No. of accessions for AAR		
	Min	Max	< 1.2	> 2.0	1.2 – 2.0
T1	0.90	4.06	4	10	14
T2	0.85	2.00	4	0	6
T3	1.31	2.4	0	5	7

Results and Discussion (2)

➢ PHS induced α-amylase activity has been reported to be much higher in the embryo (head) than in the distal (tail) half of the grain; whereas LMA-induced α-amylase was present to a similar degree in both halves (Mares and Mrva, 2008). Thus α-amylase ratio (AAR) derived from head/tail can be used to differentiate PHS and LMA.

➢ Thirty accessions showed low FN in one or more of the three treatments. Eight accessions showed AAR smaller than 1.2, suggesting LMA damage (four in T1 and four in T2). Ten in T1 and five accessions in T3 showed AAR greater than 2.0, suggesting PHS damage. The remaining 27 accessions showed AAR between 1.2 and 2.0, suggesting both LMA and PHS damage (Table 2).

➢ The AAR threshold used in this preliminary study needs to be validated with additional data as we cannot explain why five low-FN accessions in T3 showed AAR greater than 2.0 with no lodging (Table 2).

Results and Discussion (3)

➢ PCA identified three subpopulations (Fig 2) in the 123 accessions: group A was mainly from West Asia, and group B mainly from south Asia, and group C from the rest of the world. The structure of the collection is highly related to their geographic origin.

➢ Using general linear model in association mapping, 5 QTL located on chromosomes 3A, 3B, 6A, 7A, and 7D were associated with FN data derived from T1; three QTL on 3B, 6A, and 6D were associated with FN data from T2; and 2 QTL on 6A and 6D were associated with FN data from T3.

➢ Previous studies found QTL for PHS mainly on 2B, 2D 3A, 3B, 3D and 4A (Kulwal et al. 2012); QTL for LMA were identified on 3B, 6B, 7B (Emebiri et al. 2010).

➢ A validation study under LMA- and PHS-conductive conditions is needed to confirm the QTL identified in this set of accessions.

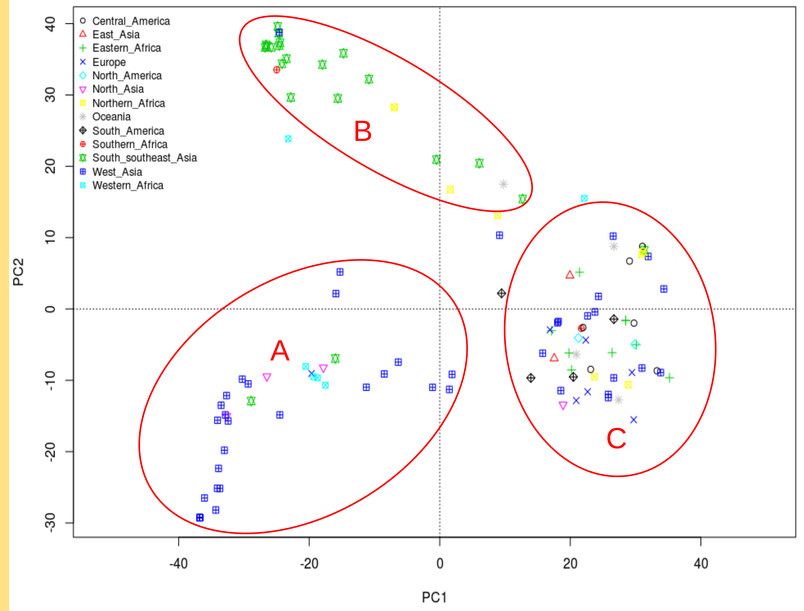

Fig 2. Principal component analysis (PCA) of the population structure in123 HWS accessions

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