

Genetic analysis of preharvest sprouting associated alpha amylase activity in barley

Rui Wang¹, Gurkamal Kaur¹, Marta Izydorczyk², Dean Spaner³, Aaron D. Beattie⁴, Ana Badea⁵, Belay T. Ayele¹

¹Department of Plant Science, University of Manitoba, Winnipeg, Manitoba, Canada ²Grain Research Laboratory, Canadian Grain Commission, Winnipeg, Manitoba, Canada

³Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Canada

⁴Department of Plant Science, University of Saskatchewan, Saskatoon, Canada

⁵Brandon Research and Development Center, AAFC, Brandon, Manitoba, Canada

Introduction

Preharvest sprouting (PHS) is defined as the germination of grains on the mother plant prior to harvest due to wet and humid summer conditions (Gao and Ayele, 2014). Barley genotypes with low level of seed dormancy are susceptible to PHS, leading to increased alpha-amylase activity and starch degradation that causes substantial grain yield and quality losses. Therefore, identifying genetic markers/genes controlling PHS related traits is crucial for developing PHS resistant barley cultivars. The main objective of this study is to identify genetic markers associated with PHS induced alpha-amylase activity in barely seeds using a genome-wide association study (GWAS) that involves diverse barley genotypes.

Materials and Methods

Plant materials and field trials: Seeds of diverse barley genotypes were grown in replicated field trials in Brandon, MB and Edmonton, AB.

Phenotyping: Barely seeds of each genotype harvested from all environmental trials were examined for alpha amylase activity using Rapid Visco Analyzer (RVA).

Genotyping: The genotypes used for the study were genotyped using a 50k genotyping array.

Genetic analysis: Analyses to determine association between phenotypic and genotypic data were performed using multiple bioinformatic tools.

Results

Table 1. Range and average values of alpha-amylaseactivity of the genotypes under different environments

Environment	Minimum	Maximum	Mean
	RVU	RVU RVU	
EDM 2022	14	196	108
EDM 2023	17	144	94
BRD 2022	6	188	79
BRD 2023	5	143	49

High degree of variation in alpha-amylase activity (RVU value ranging from 5 in BRD 2023 to 196 in EDM 2022) was observed among the genotypes under all the trial environments (Table 1).



Figure 1. Classification of barley genotypes based on RVU from all trial environments.

➢ Based on alpha-amylase activity, the genotypes are classified into three levels of PHS resistance; low (RVU < 50), intermediate (RVU ≥ 50 & < 120), and high (RVU ≥ 120) (Figure 1).

Table 2 Analysis of variance of alpha amylase activity forbarley genotypes

Source	DF	Type III SS	MS	F	P > F
Environment (E)	3	283709	14663	250	< 0.001
Genotypes (G)	159	14663	283709	13	< 0.001
G x E	463	114	1149	3	< 0.001

Our analysis indicate that activity of alpha amylase in the grains is significantly influenced by genotype, trail environment and interaction between genotype and environment (Table 2).



Figure 2. Manhattan plot of pooled sample. Significant markers are shown above the broken threshold line.

- Nine significant genetic markers explaining phenotypic variation ranging from 12.6% to 16.2% were identified by our analysis.
- All the nine genetic markers represent a specific genetic locus.

Conclusion

- The genotypes studied showed significant variation in alpha-amylase activity and thereby degree of PHS resistance.
- Nine genetic makers are found to be associated with alpha-amylase activity in barley grains.

Reference

Gao, F., Ayele, B.T. (2014) Functional genomics of seed dormancy and preharvest sprouting in wheat: advances and prospects. Frontier in Plant Science 5: 458.

Acknowledgement

