Investigating Winter Hardiness to Advance Winter Malting Barley as a Climate Adaptation Strategy in Michigan (CBC # 200000001789)

2020-21 Final Report

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Abstract

In 2020-21, our team received support from the Michigan Craft Beverage Council to investigate winter hardiness in malting barley to advance winter barley as a climate adaptation strategy. We conducted a hybrid field and laboratory study to assess the interaction of winter barley genetics with environmental conditions across locations in Michigan and their combined effect on winter barley hardiness. Results from our first year of research point to promising winter hardy varieties and the important role of snow cover in protecting the winter barley crop. Further research is needed to fully assess the mechanisms driving species/variety differences in winter hardiness, including the role of antioxidative enzymes.

Introduction

Of the crops currently produced in Michigan as craft beverage ingredients, spring malting barley may be especially vulnerable under projected climatic changes (Niero et al., 2015). The Michigan malting barley industry has identified winter barley production as a promising alternative to spring barley, which may be better adapted to the agroecology of our state and unique opportunities/risks that climate change presents. Yet, winter barley remains susceptible to winter injury, suggesting that ongoing efforts to identify resilient winter barley varieties and environmental thresholds for winter survival should be prioritized.

In 2020-21, our team received support from the Michigan Craft Beverage Council to begin investigating winter hardiness in malting barley to advance winter barley as a climate adaptation strategy. We proposed three primary objectives for the project focused on 1) an applied field study assessing the interaction of winter barley genetics with environmental conditions across locations in Michigan and their combined effect on winter barley hardiness; 2) sampling of barley tissue from the field study and laboratory analysis to measure traits associated with winter barley hardiness; and 3) development of models to predict winter barley hardiness and distribution of this new information through extension outreach. We report significant progress on each of our stated objectives below.

Objective 1: Compare winter injury and survival of eight elite winter barley cultivars under variable temperature, precipitation and snow cover conditions at six locations across Michigan representing USDA hardiness zones 4a – 6b.

Hypothesis: With a killing temperature of -10 degrees C, winter barley will experience significant cold injury most years in zones 4a – 5b. Site-year variation in temperature, precipitation, humidity and light (sun and snow cover) will correlate with observed barley injury and survival.

Five research sites were established in the fall of 2020 at Empire (6b), KBS (6a), Stockbridge (5b), UPREC (5a) and Brimley (4b), MI. Each location included seven winter barley varieties and one hardy cereal rye check planted in a randomized complete block design with 3-4 replications. Barley varieties were selected based on available information regarding their winter hardiness, targeting some hardy and some non-hardy varieties. Sensors were installed at planting to monitor temperature and relative humidity at the soil surface and snow cover (light and cameras) throughout the winter (Table 1). Stand counts were taken in fall and again after spring green-up to measure stand loss over the winter (Figure 1a). Canopy cover measurements were taken in the spring using the Canopeo app to compare relative leaf area (Figure 1b).

Location (Hardiness Zone)	Lat.	Dist. to Lake (Mi)	Plant Date	Avg. Fall Pop. (1 Ft^2)	Fall GDDs (32 F)	Min Crop Temp (F)	Snow Period	Max Snow Depth (In)
Empire (6b)	44.81	2.51	9/17	25.17	1,537.87	9.22	12/13-3/10	11
KBS (6a)	42.41	45.30	9/23	29.78	1,537.45	20.53	12/24-2/28	13
Stockbridge (5b)	42.45	56.70	11/5	29.49	454.84	21.66	12/25-2/28	16
UPREC (5a)	46.35	6.67	9/10	22.92	1,083.60	17.98	12/12-3/15	17
Brimley (4b)	46.40	1.34	9/18	17.25	1,091.34	16.32	12/14-3/18	18

Table 1: Environmental and crop conditions at the five research sites in 2020-21

This work has begun to reveal key variety differences and environmental factors driving winter barley hardiness. For example, stand loss over the winter varied significantly across varieties, by location (Fig. 1a). Only one location with limited snowfall, Empire (6b), experienced temperatures at the soil surface below the known cold threshold for barley (-10 C / 14 F). UPREC (5a) experienced significantly more, and KBS (6a) significantly less, stand loss than the other locations. While no single barley variety performed well at all locations, Thoroughbred was consistently a poor performer experiencing significant stand loss at most locations. LCS Violetta and Wintmalt each incurred the lowest amount of stand loss among varieties at two separate locations. In addition to these variety differences, environmental factors found to be significantly correlated with stand loss in our study were latitude ($t = 3.32$, $P<0.0001$), number of days with snow cover ($t = -3.02$, $P = 0.02$), and maximum snow depth ($t = -2.38$, $P = 0.019$). Together, barley variety and the environmental factors noted above explained 40% of the variation in observed stand loss. One factor that limited our ability to accurately assess stand loss in this study was tillering of individual barley plants from planting through spring green-up. This will be addressed in the coming year through destructive sampling, digging of subsample areas from each plot to better distinguish individual plants.

Spring canopy cover was correlated with stand loss ($r = -0.64$) and also varied significantly across varieties, by location (Fig. 1b). Stockbridge (5b) and KBS (6a) had significantly more, Empire (6b) and UPREC (5a) significantly less, spring canopy cover than Brimley (4b). LCS Violetta and Wintmalt each had the highest canopy cover at two of five locations. Thoroughbred had the lowest spring canopy cover at three of five locations. In addition to these variety differences, environmental factors found to be significantly correlated with spring canopy cover in our study were latitude ($t = -11.13$, $P<0.0001$), fall growing degree-day accumulation ($t = 9.98$, $P = 0.01$), and minimum

Figure 1a

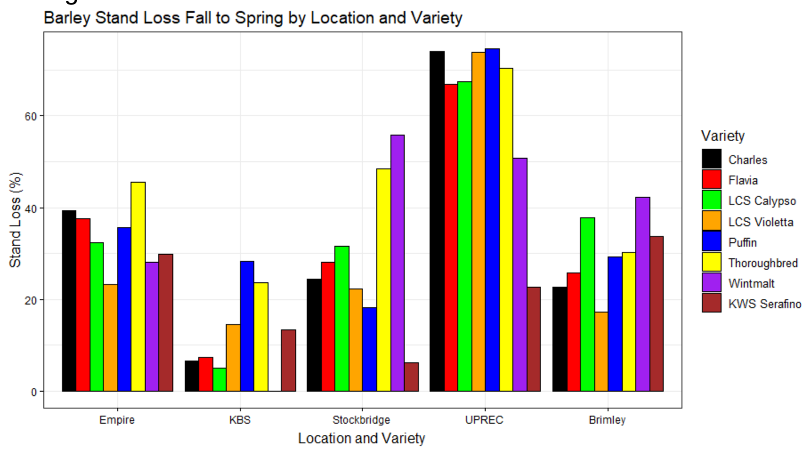
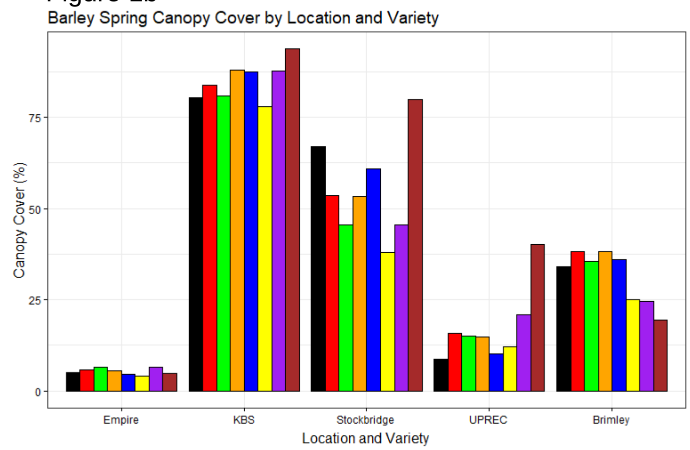


Figure 1b



winter temperature (t = 14.50, $P<0.0001$). Together, barley variety and the environmental factors noted above explained 84% of the variation in spring canopy cover. One factor that limited our ability to easily interpret spring canopy cover data from this study was inconsistent timing of canopy cover measurements across locations. This will be addressed in the coming year by timing canopy cover measurements according to barley growth stage.

Objective 2: Quantify known and novel traits previously associated with winter hardiness by sampling barley tissue from the proposed field trial and analyzing it for desiccation, sugar accumulation, fatty acid desaturation, antioxidative enzyme levels and expression of DNA repair genes.

Hypothesis: Varietal differences in winter barley cold hardiness will be observed in the field, and largely explained by the measured covariates.

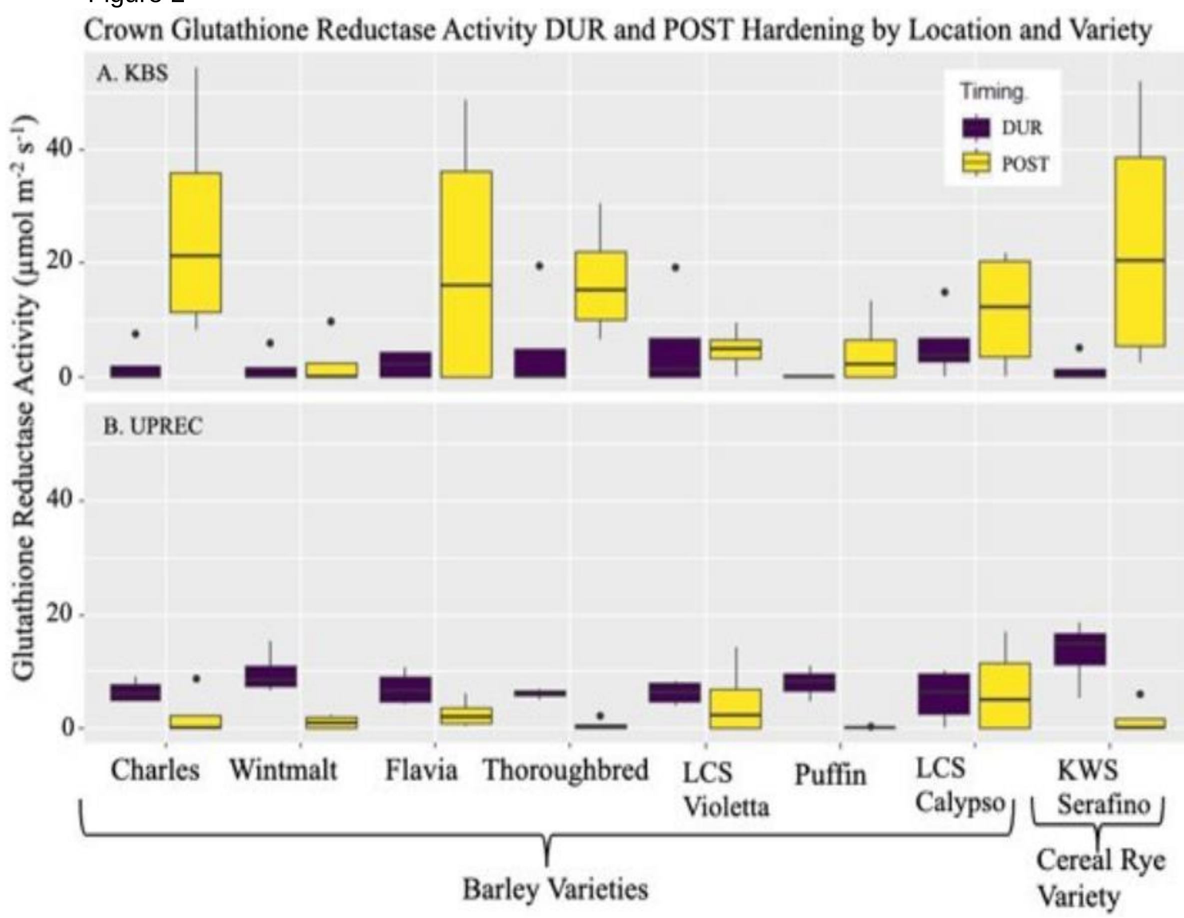
Oxidative stress (caused by buildup of reactive oxygen species) is a common feature of extreme temperature stress involving either heat or cold. Plants have a comprehensive suite of enzymes that mitigate oxidative stress by breaking down reactive oxygen species into nontoxic products. However, in order to handle extreme cold temperatures associated with a Michigan winter, plants must invest not only in producing sufficient quantities of these enzymes but also in maintaining their functionality at low temperatures. While a number of studies have documented increased production or activity of antioxidative enzymes under freezing stress in plants like wheat (e.g. Baek and Skinner, 2003), few have compared antioxidative enzyme activity under cold stress among cold-tolerant and cold-sensitive cultivars of a single species.

We focused the initial laboratory portion of our project on analysis of Glutathione reductase (GR) activity during and after cold hardening. In plants, reduced glutathione participates in the glutathione-ascorbate cycle in which it reduces dehydroascorbate, a reactive byproduct of hydrogen peroxide. In this way, GR is believed to aid plant responses to abiotic stresses like salinity, drought or extreme temperatures. Barley tissue samples were collected before and after cold hardening in the fall from two of our field locations (UPREC and KBS). Leaf and crown tissue was gathered from each plot, preserved on dry ice, and moved to the Merewitz-Holm lab at MSU for analysis.

Our study found that GR activity was significantly higher in barley leaf tissue relative to crown tissue ($P < 0.0001$). Despite higher GR levels in leaf tissue, greater differences were observed between locations and species in *crown* GR activity, highlighting the critical importance of crown tissue/conditions for winter survival. Overall, GR activity varied significantly between locations ($P = 0.04$), but not between winter barley varieties (Figure 2). However, cereal rye (KWS Serafino) had significantly higher GR activity ($P = 0.08$) than all winter barley varieties in crown tissue during hardening at UPREC. The average change in crown GR activity during to after hardening was significantly different between locations ($P < 0.0001$), increasing at KBS and decreasing at UPREC.

Our hypothesis was not fully supported, as barley varieties did not differ in GR activity. However, cereal rye showed higher GR activity than barley at one location*timing, which suggests that low GR activity could partially explain the poor winter hardiness of barley relative to rye. The strong influence of location on GR activity suggests important interactions between genetics and environment that require further investigation. Further research in controlled growth chamber environments may be necessary to fully understand the role of GR and other antioxidative enzymes in winter barley hardiness. This portion of the project is being led by Dr. Merewitz-Holm's graduate student, Megan Gendjar, and will continue into the next year as she gains experience with new equipment and analysis protocols.

Figure 2



Objective 3: Improve winter barley variety development and Michigan site selection by constructing predictive winter hardiness models and sharing this new information through extension outreach.

Hypothesis: Screening and selection of barley varieties for the most important traits identified by our work will help to improve winter hardiness long-term, while Michigan growers, maltsters and brewers will benefit from more precise recommendations regarding site selection for specific winter barley varieties.

While Michigan winters appear stark at face value (i.e. cold, dark), winter is truly a complex and dynamic phenomenon presenting multiple forms of abiotic stress at varying levels for winter cereals like barley. Our data from this first year of research suggest that 1) No one winter barley variety is suited to all Michigan environments, but rather each has unique strengths and weaknesses. That said, there are clearly "better bets" when it comes to winter hardiness, like LCS Violetta and Wintmalt; 2) USDA hardiness zone is likely a good, but imperfect, predictor of winter barley hardiness because it is based on average minimum air temperature, which we found to be important for barley growth (canopy cover), but less so for stand loss; 3) Sufficient snow cover may be one of the most important factors for winter barley hardiness. Climate change effects on snow cover resulting in less, or less predictable, snowfall from year to year could be detrimental to winter barley hardiness; and 4) Differences in antioxidant enzyme activity may explain some of the variation in winter barley hardiness relative to other cereals like rye or wheat. The fact that our study has so far failed to identify a winter barley variety that demonstrates excellent hardiness across environments indicates that our path forward may be in pairing individual barley varieties with the particular environmental conditions to which they are best adapted. However, additional observations are required to replicate and validate these initial findings from our first year of research.

To-date, information on our project and initial findings has been shared with growers and others through several outlets including the 2020 and 2021 Great Lakes Hop and Barley Conferences (115 participants), the 2022 Michigan Great Beer State Conference (362 participants), the 2021 KBS Small Grains for Brewing and Distilling field day (21 participants), the 2021 UPREC legislative tour, UP Ag Connections Newsletter (1,100 recepients), and social media including the "What's UP @ UPREC" video series (60 views). The Craft Beverage Council granted special permission to use $4,000 of grant funds to support travel of researchers to present at the 2022 Michigan Great Beer State Conference in Traverse City, MI. The conference was integral to strengthening our relationships with researchers beyond the project team who have experience in winter barley breeding and hardiness. We intend to collaborate with researchers from Cornell University in the year ahead to draw on their knowledge of this topic and experimental germplasm previously rated for winter survival in New York State. The project was otherwise conducted consistent with the budget proposed by the principal investigator and approved by the State of Michigan. We will repeat this experiment in 2022-23 with on-going support from the Craft Beverage Council, incorporating lessons learned from the results reported here.