AN ABSTRACT OF THE THESIS OF

Brigid M.A. Meints for the degree of Master of Science in Crop Science presented on June 12, 2014.

Title: From Germplasm Development to Variety Release: The Oregon State University Food Barley Experience

Abstract approved:

Patrick M. Hayes

Barley (*Hordeum vulgare* L.) is one of the oldest known domesticated crops. Originally cultivated for human consumption, other end-uses have gained importance over the millennia. Barley is the fourth most important cereal crop in the world (FAO-STAT, 2011), and today it is mainly used as animal feed or malted for brewing and distilling, while wheat and rice have replaced it as a food product. But the food barley movement is being revived in many parts of the world (Baik and Ullrich, 2008; Bhatty 1999; Dickin et al., 2012; Grando and Gomez Macpherson, 2005), including the Pacific Northwest of the US. The Oregon State University Barley Project is currently developing novel food barley varieties with interesting colors, flavors, and nutritional qualities. Our most advanced food lines were grown in the OFOOD trial: a multi-location, multi-year trial consisting of 14 experimental lines grown under dryland, irrigated, and high rainfall conditions across the Pacific Northwest. This trial

consisted of a mixture of hulled lines with waxy starch and hull-less lines with normal starch. The lines were evaluated for agronomic and food quality traits and resistance to biotic and abiotic stresses. One of the entries in the OFOOD trial is a non-waxy hull-less line called ‘Streaker’. Streaker is a blend of three sister lines that has blue, white, and brown kernels, and will be released within the next year. This thesis follows food barley research at OSU from the original breeding scheme and definition of objectives, to variety trialing and quality characterization, and finally to germplasm release and product development.

From Germplasm Development to Variety Release: The Oregon State University Food Barley Experience

by
Brigid M.A. Meints

A THESIS

Submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

Presented June 12, 2014
Commencement June 2015

Master of Science thesis of Brigid M.A. Meints presented on June 12, 2014.

APPROVED:

Major Professor, representing Crop Science

Head of the Department of Crop and Soil Science

Dean of the Graduate School

I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature bellow authorizes release of my thesis to any reader upon request.

Brigid M.A. Meints, Author

ACKNOWLEDGEMENTS

There are not enough words to express the amount of gratitude that I have for my barleyworld family. First and foremost, I must thank my major professor Dr. Patrick Hayes who changed my life when he gave me the opportunity to pursue plant breeding. Without his guidance and assistance, I would not be where I am today. Thanks to other barleyworld members and affiliates: Dr. Alfonso Cuesta-Marcos for his statistics advice, Tanya Filichkin for her lab prowess, and Dr. Andrew Ross for introducing me to the world of cereal chemistry. Thanks to Scott Fisk and Laura Helgerson for their friendship and support. Thanks to my fellow barley graduate students and officemates: Araby Belcher, Ryan Graebner, and Dustin Herb for their humor and wisdom. Thanks to my graduate council representative, Dr. James Hermes, for his valuable time and expertise.

Team food barley for life!

CONTRIBUTION OF AUTHORS

Dr. Patrick M. Hayes initiated the project and guided it along the way. Dr. Alfonso Cuesta-Marcos assisted with statistics and contributed to the manuscript. Scott Fisk managed the field trials in Corvallis, OR. Dr. Andrew Ross advised the food quality portion and contributed to the manuscript. Teepakorn Kongraksawech helped with quality analysis. Dr. Juliet Marshall and Chad Jackson conducted field trials in Aberdeen, ID. Dr. Kevin Murphy carried out field trials in Pullman, WA.

TABLE OF CONTENTS

Page

GENERAL INTRODUCTION 1

FOOD BARLEY QUALITY AND GERMPLASM UTILIZATION 4

REFERENCES 44

DEVELOPING WINTER FOOD BARLEY FOR THE PACIFIC NORTHWEST OF THE US 65

ABSTRACT 65

INTRODUCTION 66

MATERIALS AND METHODS 71

RESULTS AND DISCUSSION 74

CONCLUSIONS 83

REFERENCES 85

REGISTRATION OF 'STREAKER+' BARLEY 108

ABSTRACT 108

INTRODUCTION 108

METHODS 110

CHARACTERISTICS 112

REFERENCES 118

GENERAL CONCLUSIONS 123

BIBLIOGRAPHY 125

LIST OF FIGURES

Figure		Page
1.1	Description of food barley germplasm used in the Oregon State barley breeding program starting in 2005 with the first crosses contributing to the OFOOD trial to the DHGS program in 2014. This figure details the diverse germplasm that has contributed to the different trials described in this chapter	57
1.2	Graph showing the mean yield rankings (1 = highest yield) compared to standard deviations of the rank for the OFOOD trial over two years at six locations. Green lines indicate median value	58
1.3	Streaker Barley Flakes from Camas Country Mill (Alvadore, OR, USA)	59
1.4	Vitamin E content for the standard reference panel for 2011-12. aT = alpha-tocopherol; gT = gamma tocopherol; aT3 = alpha-tocotrienol; gT3 = gamma tocotrienol; dT3 = delta-tocotrienol. Alpha-tocopherol is the form required by humans, but all are potent lipid soluble antioxidants	63
1.5	Doubled Haploid Genomic Selection breeding scheme. Germplasm described in Fig. 1 will be included in the training population	64
2.1	Consistency plot of mean rank by standard deviation of rank for grain yield at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values	93
2.2a	AMMI1 plot for grain yield at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013	94
2.2b	AMMI2 plot for grain yield at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013	95
2.3	Consistency plot of mean rank by standard deviation of rank for height at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	96
2.4a	AMMI1 plot for plant height at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013	97
2.4b	AMMI2 plot for plant height at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013	98

LIST OF FIGURES (Continued)

Figure		Page
2.5	Consistency plot of mean rank by standard deviation of rank for heading date at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	99
2.6	Consistency plot of mean rank by standard deviation of rank for test weight at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	100
2.7	Consistency plot of mean rank by standard deviation of rank for β-glucan at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	101
2.8	Consistency plot of mean rank by standard deviation of rank for protein at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	102
2.9	Consistency plot of mean rank by standard deviation of rank for kernel hardness at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	103
2.10	Consistency plot of mean rank by standard deviation of rank for solvent retention capacity of water (SRC-W) at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	104
2.11	Consistency plot of mean rank by standard deviation of rank for winter survival at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.	106
2.12	Pearson's simple correlation coefficients and *P* values for traits measured on all 16 entries in the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013	107
3.1	Pedigree contributing to Streaker+. Varieties in bold were developed by other breeding programs. Underlined varieties were released from the Oregon Agricultural Experiment Station with the year of release in parentheses.	119

LIST OF TABLES

Table		Page
1.1	β-glucan, grain protein, and yield measurements for Streaker at three locations representing different climate conditions in 2011-12 and 2012-13.	59
1.2	Barley Breakfast Bar recipe served at the Bethel School District (Lane County, OR, USA). The recipe was adapted to include barley flakes, like those shown in Fig.1.3	60
1.3	Food barley standard reference panel for 2011-12 and 2012-13. See text for details on equipment and assays	61
1.4	Mineral content for the standard reference panel for 2011-12. Values per 100 g. USDA standard values for hulled and pearled barley listed for comparison (no data available for Mn, Cu, or B).	62
1.5	Mineral content for the standard reference panel for 2012-13. Values per 100 g. USDA standard values for hulled and pearled barley listed for comparison (no data available for Mn, Cu, or B)	62
2.1	Pedigree, row type, hull type, and starch type for all lines in the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.	90
2.2	Means of grain yield, heading date, height, test weight, β-glucan, protein, kernel hardness, and solvent retention capacity of water (SRC-W) of the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.	91
2.3	Estimates of variance components for grain yield and height for the OFOOD trial at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.	92
2.4	Means for barley stripe rust (BSR) and scald for the OFOOD trial at Corvallis, OR in 2012 and 2013. Means for winter survival for the OFOOD trial at Pullman, WA and Aberdeen, ID in 2012 and 2013.	105

LIST OF TABLES (Continued)

Table		Page
3.1	Pairwise genetic differences between each of the three components of Streaker+ based on 6,895 molecular markers on the Infinium iSelect 9K genotyping chip	120
3.2	Agronomic performance and food quality of Streaker and check varieties across 13 environments (4 high rainfall, 4 dryland, 5 irrigated).* *Corvallis, Hermiston, Lewis-Brown, and Pendleton, OR; Mount Vernon and Pullman, WA; Aberdeen and Parma, ID.	120
3.3	Agronomic performance and food quality of Streaker and check varieties across 4 dryland environments.* * Pendleton, OR and Pullman, WA.	121
3.4	Agronomic performance and food quality of Streaker and check varieties across 4 high rainfall environments.* * Corvallis and Lewis-Brown, OR; Mount Vernon, WA.	121
3.5	Agronomic performance and food quality of Streaker and check varieties across 5 irrigated environments.* * Hermiston, OR; Aberdeen and Parma, ID	122
3.6	Reaction of Streaker and check varieties to barley stripe rust and scald, and winter survival. * Based on a 1-9 rating scale where 1 = most resistant and 9 = most susceptible.	122

From Germplasm Development to Variety Release: The Oregon State University Food Barley Experience

General Introduction

Barley (*Hordeum vulgare* L.) is one of the oldest known domesticated crops. Originally cultivated for human consumption, other end-uses have gained importance over the millennia. Barley is the fourth most important cereal crop in the world (FAO-STAT, 2011), and today it is mainly used as animal feed or malted for brewing and distilling, while wheat and rice have replaced it as a food product. But the food barley movement is being revived in many parts of the world (Bhatty 1999; Grando and Gomez Macpherson, 2005, Dickin et al., 2012; Baik and Ullrich, 2008), including the Pacific Northwest of the US. In the United States, this effort is due in part to new reports that show that most North Americans do not get enough fiber in their diets (Park et al., 2011) and increasing rates of obesity, heart disease, cancer, and diabetes (Go et al., 2013). In 2006, the U.S. Food and Drug Administration approved a health claim for barley based on its potential for high levels of the soluble fiber, β-glucan, which has been shown to help reduce post-prandial glucose response, lower blood cholesterol levels, reduce insulin resistance, and reduce abdominal fat (AbuMweis et al., 2010; Bays et al., 2011; Behall et al., 2006; Casiraghi et al., 2006; Kim et al., 2009; Shimizu et al., 2008; Tiwari and Cummins, 2011). The health claim allows "foods containing barley to claim that they reduce the risk of coronary heart disease. Specifically, whole grain barley and dry milled barley products such as flakes, grits, flour, and pearled barley, which provide at least 0.75 grams of soluble fiber per

serving" (21 CFR 101.81) (Ames and Rhymer 2008; National Barley Foods Council, 2003). The Oregon State University (OSU) barley breeding project projects that with variety development and research on quality traits, barley can make a comeback as a nutritious and delicious food crop and can serve to alleviate some of these ills. This thesis serves to report our latest exciting work on food barley.

In chapter one, we present an extensive literature review on the current research being done on food barley variety development, quality characterization, and product development. We also give a brief overview of the trajectory that the OSU food barley breeding program has followed over the last decade. We introduce the Oregon Food Barley (OFOOD) variety trial, which is discussed in greater depth in chapter two, and the first winter food barley release from the OSU program, 'Streaker+', which is discussed further in chapter three. Based on the literature, we justify certain decisions that our program has made: pursuing a whole grain model rather than an extractive one, exploring new food quality traits such as kernel hardness and solvent retention capacity, and breeding for specific characteristics, including the hull-less trait.

Chapter two describes the OFOOD trial, which is the first food barley variety trial with germplasm developed by our breeding program. This multi-location, multi-year trial represents our most advanced food lines developed through marker-assisted and phenotypic selection. The goal was to create a set of winter/facultative barley lines with waxy starch that would be adapted to the Pacific Northwest. We present data for agronomic traits, grain and food quality traits, and resistances to biotic and

abiotic stresses. We look at the significance of main effects as well as genotype x environment interactions. Out of this trial, two lines were identified for release.

Chapter three is the Streaker+ germplasm release manuscript, which describes its agronomic and quality profile. Streaker+ is a winter hull-less food variety that is a blend of three pure sister lines that form an appealing mixture of blue, brown, and white kernels. Streaker is already being grown commercially with great success, and food product development featuring Streaker is currently underway.

This thesis follows food barley research at OSU from the original breeding scheme and definition of objectives (chapter one), to variety trialing and quality characterization (chapter two), and finally to germplasm release (chapter three) and product development (general conclusions).

Food barley quality and germplasm utilization

Brigid Meints, Alfonso Cuesta-Marcos, Scott Fisk, Andrew Ross, Patrick Hayes
Oregon State University, Department of Crop and Soil Science, Oregon State University, Corvallis, OR 97331, USA

In Exploration and Utilization of Barley Genetic Germplasm

Published by Jones Wiley Press and Zhejiang University

(submitted)

Introduction

Barley is one of the oldest known domesticated crops. Originally cultivated for human consumption, other end-uses have gained importance over the millennia. Barley is the fourth most important cereal crop in the world (FAO-STAT, 2011), and today it is mainly used as animal feed or malted for brewing and distilling, while wheat and rice have replaced it as a food product. But there are still many areas of the world where barley remains a staple crop and has important spiritual, nutritional, and cultural significance. There are a number of excellent reviews on food barley that have been published in the last few years (Grando and Gomez Macpherson, 2005; Baik and Ullrich, 2008; Newman and Newman, 2008; Baik et al., 2011). Therefore, in this review, we will summarize the history of barley foods, discuss their resurgence, and use the Oregon State University (OSU) food barley breeding initiative as a case study to share the current state of our food barley germplasm development, breeding targets, and breeding strategies.

A Brief History of Barley Foods

There are multiple theories regarding the location and rationale behind the domestication of barley. Archaeological evidence revealed that pre-domestication wild barley, *Hordeum vulgare subsp. spontaneum*, was consumed as early as 17,000 years BCE in the Fertile Crescent (Newton et al., 2011). Traditionally, barley is said to have arisen from a single domestication event in the Fertile Crescent (Nevo, 1992) but recent evidence suggest that there may have been multiple domestication events

in the Fertile Crescent and Central Asia or Africa (Vavilov, 1951; Dai et al., 2012). Morrell and Clegg (2007) used haplotype segments from a distribution of wild barleys to show that allelic composition differed based on geographic location, leading them to conclude that there were two separate domestication events: one in the Fertile Crescent, and another 1,500-3,000 km to the east.

Domestication took several thousand years to complete, with the most noticeable changes being the non-brittle rachis, which prevented the spike from shattering, increased seed weight and plumpness, the selection (in some areas) of six-rowed spikes, and free-threshing grain (Salamini et al., 2002). Selection for the hull-less caryopsis (the phenotype where the lemma and palea do not adhere to the hull, sometimes referred to as 'naked') was particularly relevant in areas where barley is consumed directly as a food. Based on allelic analysis, Taketa et al. (2004) concluded that the hull-less trait was the result of a single domestication event, most likely in southwestern Iran. The role of the adhering hull (controlled by the *Nud* gene) (Taketa et al., 2008) is particularly important in defining end use: hull retention is very important for malting and brewing, where the hull serves as a natural filter during the brewing process. As an animal feed, hulls may or may not have value. In the case of human food, the hull has little value as it consists of insoluble fiber (Baik et al., 2011) and for maximum palatability and ease of processing it is removed by pearling or de-hulling.

In the popular imagination, the domestication of barley is often associated with the invention of beer (typified by the Discovery Channel documentary film

"How Beer Saved the World"). In fact, barley foods and beers were likely developed simultaneously and the two were indistinguishable: beer was food and food was beer. Archaeological data suggests that in the Fertile Crescent, barley was consumed both as an alcoholic drink and a fermented dough that was ground into meal and mixed with spices (Newman and Newman, 2008). Cultivation of barley for human consumption began approximately 10,000 years ago and the crop eventually became a staple food for a diverse set of cultures around the world, each developing unique preparation processes, recipes, and methods of consuming the grain. On the whole, barley foods were for the poor. One of the tastier tidbits of barley food history is that the Roman gladiators were called the "hordearii" (barley men) because they subsisted on barley bread. Scottish peasants, whom we may associate more with oats than barley, apparently subsisted – morning, noon, and night – on barley porridge, with a side of boiled greens on a good day. In Tibet, barley was, and continues to be, an essential part of the daily diet. Toasted barley flour, along with green tea and yak butter, is used to make a food called tsampa. Special barley foods continue to be popular in some regions and may have unique health-promoting properties. For example, "dakos" is a traditional barley rusk on the island of Crete. Local bakeries use flour milled from whole (hulled) barley. In some cases, even the awns are ground with the grain. This whole grain barley food, awns and all, is thought to be one of the reasons for the low rate of colon cancer in the Cretan region

Not all barleys used for human food are hull-less. However, the prevalence of hull-less barley types is highest in regions where barley foods were, and remain,

important staples – Ethiopia, the Himalayan region, China, Korea, and Japan. Barley is an important food in the Andean region of South America; the first barleys introduced into this area with the Spanish conquest likely had hulls, but there are also hull-less Spanish barley landraces that may have been introduced at the same time.

The Renaissance of Barley Foods in Western Culture

Despite its rich cultural and culinary significance in many cultures around the world, in many modern day societies, barley has all but disappeared as a food raw material despite its virtues as a fiber-rich and versatile grain. Creating a food barley market in the 21st century has proven as challenging as creating a market for entirely novel grains, such as teff or quinoa. However, there is renewed enthusiasm for food barley and it comes from increasing public awareness of the value of healthy eating. To support the value of barley as part of mainstream diets we cite the AARP-NIH cohort study (Park et al., 2011). In this study of 388,000 participants, dietary fiber was linked to decreased risk of death from cardiovascular disease, cancer, and infectious and respiratory diseases. Notably, Park et al. (2011) concluded that "Dietary fiber from grains, but not from other sources, was significantly inversely related to total and cause-specific death in both men and women". This conclusion emphasizes the need to get food products rich in cereal fiber into mainstream diets. Barley, as a rich source of cereal fiber and other phytonutrients as a whole-grain (Jones, 2010), is part of the solution and can help to address one of the world's emerging health challenges: the grossly inadequate fiber intake of most North

Americans and, by extension, many urban dwellers around the world (Slavin, 2005). The principle fiber found in barley is β-glucan, a soluble fiber. Barley has the advantage for consumers as it provides its fiber and other healthful components in a package that has half (or less) of the fat content of the other main cereal β-glucan source, oats (Svihus and Gullord, 2002), and with a greater total dietary fiber content than wheat, oats, or rye (Cho et al., 1999; Izydorczyk, 2010). Barley β-glucan is effective in reducing the incidence and severity of "metabolic syndrome" (PubMed Health, 2011) through increased satiety, slowed macronutrient absorption, reduced post-prandial glucose response, lowered blood cholesterol levels, reduced insulin resistance, and reduced abdominal fat (AbuMweis et al., 2010; Arndt, 2006; Bays et al., 2011; Behall et al., 2006; Casiraghi et al., 2006; Kim et al., 2009; King et al., 2008; Shimizu et al., 2008; Thondre and Henry, 2009; Tiwari and Cummins, 2011;Vitaglione et al., 2010). The capacity of barley foods to reduce cholesterol was the key factor in the successful approval of the FDA health claim for barley in 2006 (21 CFR 101.81) (Ames and Rhymer, 2008; National Barley Foods Council, 2003). There have been similar health claims approved for barley in Europe in 2011 (EFSA Journal 2011, 9 (12): 2471) and Canada in 2012 (http://www.hc-sc.gc.ca/fn-an/label-etiquet/claims-reclam/assess-evalu/barley-orge-eng.php) as well. Barley also supplies other bioactive nutrients (phenolics, phytate, and tocols) that are potent antioxidants (reviewed by Baik et al., 2011; Holtekjolen et al., 2011). Barley can enrich foods that are otherwise lacking in these valuable components (Verardo et al., 2011).

Barley starches vary in their amylose content (Lagassé et al., 2006). Amongst starch variants, *high amylose starches* are favored for the creation of one form of resistant starch, which is formed by amylose retrogradation (also called recrystallization)(reviewed by Ross, 2011; Ross, 2013). However, even normal barley starches tend to have a higher amylose to amylopectin ratio than wheat and accordingly retrograde more readily (van Amelswoort and Westrate, 1992; Sullivan et al., 2013). Resistant starch is not digested in the human digestive tract but is fermented in the colon (Topping and Clifton, 2001; Nugent, 2005). All colonic fermentations produce short chain fatty acids but resistant starch is associated with higher levels of butyric acid (Brouns et al., 2002; Champ, 2013). Butyric acid is believed to act as a cell growth regulator and has protective effects against the onset and proliferation of colo-rectal cancers (Fung et al., 2012). High amylose barley has been incorporated successfully into foods made with composite barley/wheat flours (Hatcher et al., 2005; Lagassé et al., 2006).

A notable advantage of food barley is that it can be produced, transported, stored, and processed with currently available grains infrastructure, thereby greatly reducing the need for additional investments throughout the value chain. For consumers, barley easily fits into familiar products. It can be used as whole (intact) or cracked grain, including its use as a high-fiber and tasty rice alternative (Edney et al., 2002; Gray et al., 2010). Barley flour or its fractions can be used as components in flatbreads (Izydorczyk et al., 2008), tortillas (Prasopsunwattana et al., 2009), and even in risen breads and sponge cakes where it can provide desirable textures and

improved keeping quality (Gupta et al., 2008; Skendi et al., 2010). Breads have also been made from 100% barley flour without the admixture of wheat (Kim and Yokoyama, 2011b; Kinner et al., 2011). Adding barley to mainstream diets will add diversity of flavors, colors, and aromas as well as increasing the diversity of cereal fiber sources. The latter is important as humans need fiber from various sources for optimal functioning: e.g. β-glucan from barley and oats, arabinoxylan from rye and wheat, and pectin from fruits among others.

The Oregon State University Case Study

The Oregon State barley breeding program, like many barley breeding programs around the world, has historically focused on breeding malt and feed varieties. However, with the United States Food and Drug Administration (US-FDA) health claim and increasing research being conducted on the benefits of barley consumption in humans, our program began breeding food barley. In 2003, the first crosses designed to result in food varieties were made. At that time, the goal was to breed high β-glucan varieties (pursuant of the "extractive model" described below) with good agronomic performance. The object was to maximize the per hectare production of β-glucan.

Grain β-glucan is a quantitative trait, with several known QTL contributing to high β-glucan (Islamovic et al., 2013). Additionally, there is a pleiotropic effect of the recessive allele at the Waxy (*WX*) locus encoded by granule-bound starch synthase 1 (*GBSS1*) (Patron et al., 2002; Islamovic et al., 2013), with positive correlations

between β-glucan and waxy starch reported (Szczodrak et al., 1992; Xue et al., 1997; Wood et al., 2003). Therefore breeders have had success targeting the recessive (waxy, high amylopectin) allele at the *WX* locus to breed for high β-glucan barley. This means that many breeding lines with high β-glucan also have waxy starch. In one of OSU's first food barley screening initiatives, Rey et al. (2009) grew a set of 33 spring cultivars and advanced lines with a diverse profile of β-glucan content in dryland conditions in northeastern Oregon, USA to determine the commercial potential of this germplasm in this area. Briefly, these entries comprised a diverse set of genotypes from a number of different breeding programs featuring a combination of hulled and hull-less lines, with nearly all being waxy starch types. We found significant differences between entries for all traits. Grain β-glucan content was found to be relatively constant across locations and years, with the largest difference being genotype. With this germplasm and set of environments, genetics were more important than environment in determining grain β-glucan. Other sources suggest that environment may play a larger role when water or nutrient stress occurs (Bendelow, 1975; Savin et al., 1997).

In Rey et al. (2009), we also concluded that the hull-less trait plays an important role in yield. This is due in part to the lack of the weight of the hull, which can account for up to 13% of the total weight of the seed, and in part due to the fact that in North America, most breeding programs have put their effort into breeding malt types and have not spent much time developing hull-less lines with high agronomic value. Additionally, the embryos of barley are on the surface of the grain,

and are as such easily damaged, resulting in poor stand establishment and vigor. We found that spring growth habit hull-less waxy cultivars have severe production issues for stress-prone dryland environments and the currently available germplasm is not agronomically vigorous enough to warrant commercial production. We recommended that barley producers in dryland areas grow spring growth habit waxy hulled cultivars if they are interested in food barley production.

After this initial assessment of spring habit varieties and experimental germplasm was conducted, we turned our focus towards breeding new food barley lines with winter and facultative growth habit, since winter precipitation patterns prevail in our target environments. All available food barleys in the United States at that time had waxy starch and were spring types. Winter and facultative varieties have agronomic advantages and do not require irrigation, making them appealing to growers in the Pacific Northwest of the USA. Given the generally more optimum moisture regimes present under fall-sown conditions in this area, we reasoned that waxy (and non-waxy) types could be commercial prospects, particularly if the germplasm had sufficient winter hardiness. Briefly, in Chutimanitsakun et al. (2013) we used a marker-assisted selection (MAS) program to efficiently select for waxy starch and low temperature tolerance (LTT) at the *WX* and *VRN-H2* loci, respectively.

The rationale for selecting recessive alleles at *GBSS1* in order to increase grain β-glucan has already been described. The rationale for selecting for the winter allele at *VRN-H2* was that vernalization sensitivity can enhance low temperature tolerance (LTT) by delaying the vegetative to reproductive transition (Szucs et al.,

2007). The parents selected for this project had a range of phenotypes: Luca (two-row, normal starch, hulled, with winter growth habit, accessed from the Martonvasar Research Institute in Hungary), Merlin and Waxbar (two-row, waxy starch, hull-less, with spring growth habit, developed by Westbred) (www.westbred.com), and Strider (six-row, normal starch, hulled, with winter growth habit, released by the Oregon Agricultural Experiment Station in 1997) (http://washingtoncrop.com/documents/Barley/6-Row/Strider.pdf). A genome-wide association study (GWAS) was performed on the lines developed through MAS, as well as an additional set of non-waxy hull-less lines selected using phenotypic selection (PS). The parents in the PS panel also represented a range of phenotypes: Strider (see above), Doyce (six-row, hull-less, with winter growth habit, developed at Virginia Polytechnic Institute) (Brooks et al., 2005), Maja (six-row, hulled, with facultative growth habit, released by the Oregon Agricultural Experiment Station in 2006), and Legacy (six-row, hulled, with spring growth habit, developed by Busch Agricultural Resources Inc.) (http://anheuser-busch.com/). All accessions, and check varieties, were grown in one dryland and one high-rainfall location over multiple years and subsequently phenotyped for grain β-glucan, LTT, and vernalization sensitivity (VS), a potential component of LTT.

The lines were genotyped using a 3072 single-nucleotide polymorphism (SNP) panel with allele-specific primers. Genotyping revealed that all MAS-derived lines were homozygous dominant at *VRN-H2*, and all but one were homozygous recessive at *WX*, indicating waxy starch (Chutimanitsakun et al., 2013). The PS lines

all had normal starch with winter alleles at *VRN-H1*. Grain β-glucan percentage ranged from 5.0-7.0% and 4.1-6.3% in the waxy lines and 3.5-5.0% and 3.0-4.5% in the normal starch lines at the dryland and high-rainfall locations, respectively. Although MAS achieved the target allele at the *VRN-H2* locus, there was unexpected variation at *VRN-H1/FR-H1* and *VRN-H3* that had unexpected effects on LTT and VS. The authors confirmed that by selecting for the recessive allele at the *WX* locus, they could effectively raise the levels of grain β-glucan, (as previously reported by Xue et al. (1997)) and raised questions about the effects of location and climate on grain β-glucan percentage.

From the panel of MAS and PS developed lines, 14 were selected for further testing in an advanced yield trial. This trial became known as the 'OFOOD' trial and was grown at multiple locations over two years. Combining seven waxy hulled lines and one waxy hull-less line from the MAS-derived lines with three hull-less normal starch lines from the PS-derived lines, as well as three other hull-less non-waxy lines from other breeding efforts, and two checks (one malt, one feed) this trial represented the most advanced food germplasm from the OSU barley project (Fig. 1.1). In 2011-12, the trial was grown at eight locations, under high rainfall, dryland, irrigated, conventional, and certified organic conditions. In 2012-13, the trial was grown at six of the eight previous locations. We chose to analyze only three locations over the two-year period in order to represent each of the production systems. Phenotypic data were collected on an array of agronomic and quality traits. Grain β-glucan was measured on all lines at all locations in order to determine genotype by environment

interactions for the trait. Kernel hardness was measured on all lines, and a strong environmental effect was noted. All lines had excellent resistance to stripe rust (*Puccinia striiformis* f. sp. *hordei*), a disease that is prevalent in the area. Yield data are presented in a consistency plot, showing rankings compared to standard deviations for all locations over both years (Fig.1.2).

After considering the processing market, it was determined that although the hulled lines currently offer higher yields, for human consumption the hull-less trait is necessary unless pearling becomes a more viable option. Pearling is the act of abrading the kernel to remove the hull and outer bran layer to reduce cooking time and make the grain more palatable. Despite the desires of some cultures to produce breads and consume lines with the hulls on, in western cultures, malt varieties are the only barleys where the hull is a requirement.

The hull-less winter food barley Streaker was the one entry from the OFOOD trial that was released as a variety. Streaker is blend of three pure lines (OR85, OR86, and OR911) and has an appealing palette of grain colors: blue, brown, and white. Another line from the OFOOD trial, 09OR-86, which has superior disease resistance and threshability will be added in to the blend in 2014 to increase the heterogeneity and create an ever-evolving mixture that will fit under the name Streaker. Streaker, as a blend, will be released as a germplasm, meaning that there are no intellectual property, licensing, or plant-back restrictions. The OSU breeding program decided to advance Streaker in an evolutionary participatory breeding scheme to appeal to organic growers.

Organic growers need varieties developed specifically for organic conditions (Wolfe et al., 2008). Most varieties grown by organic farmers were bred under (and for) non-organic production conditions. As a consequence, these varieties may require improvement for one or more of the following traits: disease resistance, weed competition, input use efficiency, flavor, and nutritional quality. The wide range of diversity found on organic farms makes targeted regional breeding especially important for organic crops. One way to make the organic breeding process more relevant and effective is to use an evolutionary-participatory breeding scheme. Participatory plant breeding (PPB) is defined as the contribution of multiple participants (in this case breeders and farmers) to the selection process (Wolfe et al., 2008). An evolutionary-participatory breeding (EPB) model emphasizes the contribution of human selection combined with natural selection at site-specific locations (Murphy et al., 2005). An EPB method involves increasing genetic diversity by growing a heterogeneous population that will be better able to deal with pests and disease (*as reviewed by* Murphy et al., 2005). In the case of cereal grains (e.g. barley, wheat, and oats), varieties breed true (they are *homozygous* in genetics parlance). A key difference between a conventional variety and an EPB-derived variety is that the latter is a mixture of pure lines (*heterogeneity* in genetics parlance). Heterogeneity (e.g. diversity) is a positive attribute as it can provide buffering against changes in the environment and changes in both type and strain of crop pests. The only condition is that the crop variety must be sufficiently uniform for management and processing purposes. Our project is an excellent candidate for the organic EPB model because it

involves breeders and farmers working together to make selections based on the specific needs of the farmers, and it focuses on a heterogeneous blend of four lines that will help bolster the crop against disease and pest pressures.

Based on a genetic analysis involving 6,895 molecular markers on the Infinium iSelect 9K genotyping chip, two of the three components are nearly pure lines (OR85 and OR86 are 99.9% homozygous) whereas OR911 is 92.7% homozygous. The same analysis reveals that the pairwise genetic differences for the three varieties range from 12 to 20%. 09OR-86 is in the queue to be genotyped with the iSelect 9K chip. Therefore, our breeding scheme will capitalize on the heterogeneity present among the three genotypes as well as the heterozygosity in OR911. All three components are similar in plant height and maturity and all have a soft kernel texture. Accordingly, the Streaker blend is sufficiently uniform for production and processing.

We have initiated research on Streaker nutritional traits, processing characteristics, and product development. Our data are available at barleyworld.org/food/standard-panel. Briefly, Streaker has a grain β-glucan content of 4.3%, protein content of 12.3%, and yield of 6513 kg/ha averaged over three locations grown throughout the Pacific Northwest in the 2011-12 and 2012-13 crop years (β-glucan data for 2011-12 only). More specifically, these traits can be broken down by location into high rainfall, irrigated, and dryland areas. In Table 1.1 we present the β-glucan, protein, and yield data from these different growing conditions.

The health claim approved by the FDA allows "foods containing barley to claim that they reduce the risk of coronary heart disease. Specifically, whole grain barley and dry milled barley products such as flakes, grits, flour, and pearled barley, which provide at least 0.75 grams of soluble fiber per serving" (21 CFR 101.81). Based on the average β-glucan content in Streaker, this would mean that in order to receive the daily recommended soluble fiber, a person would have to eat at least 17g of steamed grain or 44g of bread made with 40% barley flour per serving. This amounts to a small side dish of steamed grain or only two slices of bread.

Products: A decision to embrace a whole-grain rather than an "extractive" model

There are two potential routes for incorporating the benefits of barley into the diets of consumers. One route is to develop and deploy foods made with whole-grain barley, either as flour or meal, or as more or less intact, cracked, or flaked seeds. The alternative is to fractionate by various means parts of the barley seed that are relatively enriched in the components of interest, such as β-glucan. In the OSU case study we chose to embrace the former route and develop desirable barley foods using the entire caryopsis, in the case of hull-less varieties, or minimally pearled caryopses in the case of hulled varieties. This section outlines our rationale for this decision.

One practical reason for our decision to embrace the whole-grain route is the number of research groups actively working to develop and deploy foods containing barley fractions enriched in β-glucan, or with partially purified β-glucan (e.g.

Canadian Grain Commission Grain Research Laboratory, Izydorczyk and Dexter, 2008; Agriculture and Agri-Food Canada, Ames et al., 2006). Additionally, our research facilities were less able to perform the pilot scale fractionations required for product development. A further practical consideration regarded location and timing. Our location on the U.S. West Coast places us at a time and place where there is resurgent interest in local and regional agricultural production-processing-consumption models (Cascadia Grains Conferences 2013, 2014). Local and regional grain systems are more responsive to the whole-grain message across all cereal and pseudo-cereal crops, not only barley, and we wish to leverage this interest to the advantage of food barley development. Accordingly, the development and deployment of foods based on whole-barley is a key strategy. We have taken the view that to gain a beachhead for barley in food formulations the newly reinvented ancient crop needs advocates and a higher level of visibility. We feel these aims are best achieved by the use of whole-grain barley and giving whole-grain barley prominence on labels and other promotional materials.

The extractive model is not without its merits and adherents and there is clear evidence of the health benefits to be gained from the inclusion of more or less refined barley β-glucan fractions in foods (Keogh et al., 2003; Biorklund et al., 2005; Keenan et al., 2007; reviewed by Fastnaught, 2010). Indeed, in an era where the U.S. diet is flagrantly deficient in fiber (Slavin, 2005), any method of increasing population-wide fiber intake is to be applauded. However, Brennan and Cleary (2005) cautioned about the potential effects on functionality related to the extraction of β-glucan with

potential reductions of molecular weight during the extraction process. Brennan and Cleary (2005) also cautioned about the cost of extraction procedures.

An evidence-based rationale for the focus on whole-barley rather than extracted fractions was the suggestion that the "co-passengers" in whole-grains (phenolics, waxes, minerals, vitamins, phytates, among others) may be as important as the fiber, or at least may act synergistically with the fiber components (Jones, 2010; Fardet, 2010; 2013; Fardet and Rock, 2013; Slavin et al., 2013). Further support for our approach comes from direct evidence showing that whole- and pearled-barley products are associated with improved health outcomes (Li et al., 2003; Behall et al., 2004a; 2004b; Hinata et al., 2007; reviewed by Fastnaught, 2010). Focusing on whole-barley also gives us the opportunity to leverage the potential health and culinary advantages of colored barley genotypes emanating from the OCOLOR nurseries (see below). Fardet and Rock (2013) have further suggested that the food matrix is critical to understanding micronutrient and phytochemical bioavailability. A holistic approach to understanding the impact of whole-grain consumption will build upon the substantial foundation established by reductionist approaches that investigated individual components. Establishment of a viable whole-barley food system is part of a holistic approach. The confluence of the factors noted above are the basis of our decision to embrace a whole-grain rather than an extractive model in our efforts to deliver desirable, healthful, and affordable barley-based foods to our community and beyond.

Product Development

The development and deployment of barley-based food products has been the subject of considerable activity in the scientific literature. This activity is a result of the work of multiple research groups worldwide trying to drive demand for food barley: either whole-grain or as a fiber-enriched ingredient (reviewed by Baik and Ullrich, 2008; Fastnaught, 2010; Sullivan et al., 2013). The variety of products is striking but is largely based on centuries- if not millennia-old templates for cereal-based foods. The basic templates can be synergized with modern food processing practices, and recent advances in knowledge of cereal component functionalities and interactions provide a means to achieve even more palatable and nutritious outcomes. Potential food-barley applications include simple intact-kernel applications (e.g. taking the place of rice or other grains in pilafs, porridges, stews, risottos, etc.) using entire, flaked, rolled, or cracked forms. Pearled forms are and can be used too (e.g. Risgaard, 2012). Interestingly, light pearling can increase the concentration of soluble β-glucan as a function of partially removing outer layers that are less rich in β-glucan (Zheng et al., 2000). Barley can also be utilized as flour or meal in a more or less refined form for applications in risen breads, flat breads, cakes, muffins, pancakes, noodles, and pasta among others.

It should be easy to adapt barley for applications where it is deployed in its intact, flaked, rolled, or cracked forms. However, our experience with "Streaker" showed that deployment as flakes, at least, was not necessarily straightforward. It took recognition by co-author Meints that Streaker's softer texture interacted with the

flaking process more effectively than harder textured varieties. This recognition facilitated the successful deployment of a high quality flaked product that had maximal retention of whole-caryopsis flakes and minimum breakage and powdering into flour. This thinking can be extended to considerations that there may be optima for kernel hardness, amylose content, fiber concentrations, and primary processing that lead to optimal cooked texture and acceptable cooking qualities even when barley is deployed in its simplest form as an intact-grain (e.g. Gray et al., 2010).

Flour-based applications are generally more challenging than intact-grain applications: risen (high volume) breads are probably the most challenging application. Baik and Ullrich (2008) reviewed a number of studies that investigated composite wheat/barley flour breads. Many breads were considered "acceptable", but common faults were reduced volume, darker color, harder texture. Sullivan et al. (2013) also reflected these generally negative changes in bread attributes after barley or barley fiber addition. Kinner et al. (2011) reported acceptable end-products from an "optimized" formulation that included sugar and fat. Kim and Yokoyama (2011b) reported positive outcomes for a 100% barley bread formulation using hydroxypropyl methylcellulose. We have successfully produced 100% barley-flour bread based on the template of the dense 100% rye sourdough breads of Northern Europe (e.g. Vollkornbrot and Danish rye bread), but these have been even more dense and challenging from a culinary viewpoint than their 100% rye counterparts. In these breads we used the inclusion of intact barley kernels partly for texture enhancement, but also as source of occluded (RS1) resistant starch and un-degraded high molecular

weight β-glucan. We did this partly from the caution suggested by Kim et al. (2011a) and Tiwari et al. (2011) among others about reductions in β-glucan content and molecular weight during bread processing. This phenomenon is likely accelerated in acidified breads made with sourdough (Rieder et al., 2012). We were cautioned because literature shows that physiological function of β-glucan is molecular weight, and hence viscosity, dependent (Wood et al., 2000; Tosh et al., 2010; Wolever et al., 2010). However, some processes have been shown to improve β-glucan extractability, for example in a flapjack (biscuit) (Robertson et al., 1997). These authors suggested that β-glucan exists in cells walls as a proteoglycan complex, and showed that β-glucan extractability is also enhanced with proteolysis of the food matrix. This suggests that sourdoughs, despite the risk of acid hydrolysis of β-glucan, might enhance β-glucan extractability as a result of enhanced proteolysis both by lactic acid bacteria (Di Cagno et al., 2002; Gänzle et al., 2008) and activation of endogenous cereal proteases at reduced pH. We have devoted significant time and resources to developing barley-based breads, very often with Streaker as the source of flour. We have had particular success baking breads made with between 30-70% barley flour, either yeast or sourdough leavened. The best products in our estimation so far are the tortillas, pita bread, and pretzels, commonly made with 50% barley flour. Based on informal sensory analysis, the breads, pita breads, pretzels, and tortillas have received positive responses from a number of different consumers, including farmers, millers, and professional bakers among many others.

Barley-wheat composite flours have also been investigated for noodle production. Baik and Czuchajowska (1997) used up to 20% barley flour milled from hull-less varieties. They observed little change between the mechanical properties of the cooked noodles with non-waxy barley flour and the wheat-flour control. Use of waxy barley softened noodles, which could an advantage for certain noodle types: e.g. Udon. The Wheat Enzymes and Asian Foods Laboratory at the Canadian Grain Commission (Winnipeg, Canada) has been particularly active in assessing the incorporation of barley or barley fractions into noodle products. Hatcher et al. (2005) studied pearled and roller-milled hull-less barley flours at 20 and 40% additions in composite barley-wheat flours for alkaline noodle production. At 40% barley, doughs required more water additions and noodles required shorter cooking times. The waxy flours reduced optimum cook time the most: from 6.5 to 3 minutes. The shorter cooking times were considered responsible for reducing cooking losses. Izydorczyk et al. (2005) reported similar results. Alkaline noodle color was affected "detrimentally": i.e. the noodles were darker, redder, and less yellow at 40% barley addition (Hatcher et al., 2005). There was a parallel study on dried salted noodles (Lagassé et al., 2006). In general the results for cooking times, cooking losses, and cooked noodle physical properties paralleled those seen in their prior study on alkaline noodles. The shorter optimum cook times bring up an important issue regarding the health benefits of the added barley: what happens to soluble β-glucan during cooking. It appears that the reported shorter cooking times are an advantage and that there were only small losses, in the order of 2 to 4% of total β-glucan before

cooking. Low β-glucan losses were also observed when cooking dried noodles, which necessarily take longer to cook (Izydorczyk et al., 2005).

There are other issues. Co-author Ross has ongoing concerns with what the "gold standards" ought to be when assessing barley-based breads, or other flour-based products. Should the gold standard be the refined wheat-flour product (e.g. white sandwich bread) or might a composite rye/wheat bread be a better frame of reference and standard to assess a composite barley/wheat bread. The issue of an appropriate standard in relation to noodles was discussed by Ross (2013), where the comments of Hatcher et al. (2005) were highlighted: "*While color and appearance generally play an important role in consumer acceptance and choice of food, certain food markets are more open and skewed toward less conventional products. For example, the traditional buckwheat noodles of Japan (soba) and Korea (naengmyon), deviate significantly from the common bright yellow or white color, but offer highly desirable texture, taste, and nutritional values and therefore are well established in their respective marketplaces*". We would contend that barley enriched noodles are best compared to soba rather than say bright creamy Udon made with a highly-refined white wheat flour (Crosbie and Ross, 2004). Similar comments can be made for bread and the choice of the product to use as a control can impact conclusions regarding acceptability of the barley product.

Another product under development using Streaker is an entirely barley-based injera. Injera is a traditional Ethiopian fermented flat bread typically made with teff. However, given the high prices of teff, barley is often added into the mixture,

although it is considered to be an inferior ingredient. A study published by Abraha et al. (2013) looked at the effects of different barley genotypes on injera quality and taste, using a sensory panel to judge flavor, mouth feel, texture, top surface gas holes, color, and suppleness. The authors found that genotype had a significant effect on the quality of the injera. They determined that varieties with waxy starch were unsuitable for making injera because they caused the dough to become too sticky, have a sour taste, and create too few gas holes. At Oregon State University, Solomon Yilma, of Ethiopian origin, has spearheaded research using barley for injera. Using 100% Streaker barley flour, with no added teff, and a barley starter, he has produced injera with excellent quality and taste.

Based on the initial commercial production of Streaker in the Willamette Valley of Oregon, USA in 2012-13, 10 metric tons were processed into flakes and were offered for sale by Camas Country Mill (http://camascountrymill.com/) located in Alvadore, Oregon. The Streaker flake label is shown in Figure 1.3. The Bethel School district (Lane County, Oregon) has developed a recipe for breakfast bars (Table 1.2) to accommodate the barley flakes and has had a great response from students.

The OSU barley project and the Oregon State University Food Innovation Center (http://fic.oregonstate.edu/) are currently developing a number of barley snack products using Streaker as a model. The goal is to provide a great way to get a whole grain serving and protein, while supporting Oregon agriculture. The products will be an all-natural twist on traditional granola, breakfast bars, and snack mix using

agricultural products and food ingredients that Oregon State University has developed, researched, and cultivated into producible food ingredients. The target audience will be hungry college students looking for an indulgent breakfast or snack item that is local, natural and sustainable, and OSU visitors looking for a delicious local food to take home with them.

The first step towards food barley grain availability justifies expanded product development and recipe dissemination. Once the formulation, packaging and shelf-life studies have been completed on newly developed barley products, information will be presented to entrepreneurs, farm-to-school programs, and the OSU branded products program. We are also converting existing barley recipes developed at OSU to standard formats suitable for at-home use, industrial users, and/or the USDA Nutrition Standards for School Lunch guidelines so that the recipes can be immediately integrated into institutional kitchens. The recipes will be made available via websites and electronic media maintained by the project participants, provided to school district food service managers, and shared with Oregon grain, baking, and food processing industries.

Quality Evaluations

One of the impediments to general acceptance of barley as a raw material by food processors is the lack of a classification system or specification framework. The need stems from barley's wide genotypic variation in processing and compositional traits: kernel hardness; hull-less/hulled character; starch amylose content; β-glucan

content; pericarp pigmentation; and total phenolic, phytate, and tocol contents (Baik and Ullrich, 2008). Despite the diversity of available traits at present, the commodity is generally sold only as undifferentiated “food barley”. A classification system would let buyers know what they are getting and enable breeders to target specific classes, making breeding efforts more effective. Without a workable classification system costly errors will occur: e.g., the inadvertent use of a proanthocyanidin-containing genotype, which may add color to products when it is not desirable (Quinde et al., 2004; Quinde-Axtell et al., 2005). Likewise, a soft kernel type in a pearling operation for a rice substitute may not be appropriate. Hard kernels are preferred as they pearl with minimal loss of endosperm. Delivery of the wrong type to a processor with specific raw material requirements can be a costly error (Baik and Ullrich, 2008).

We believe the above scenario is avoidable. Another project that the OSU barley project has been pursuing is a characterization of food barley quality. Because food barley has a relatively small market in the United States, there is no set of quality specifications that breeders and farmers must meet. For example, there is no analog of the hard/soft, red/white, winter/spring classification scheme like there is for wheat in the USA. We developed a Food Barley Standard Reference Panel of seven diverse varieties-- five food, one malt, and one feed. Grown for two years at various locations around the Willamette Valley of Oregon, these lines have been characterized for a range of traits. We began with tests that are typically run on all food material: a grain protein (NIR Spectroscopy; Infratec 1241 Grain Analyzer,

Foss, Laurel, MD) test and grain β-glucan assay (AACCI Method 32-23.01; Megazyme Kit, Megazyme International Ireland Ltd.) (Hu and Burton, 2008). We looked at tests typically performed on wheat and rice for guidance on tests that would be appropriate for setting quality parameters for barley. Kernel hardness was one of the first tests we chose to run, using the Single Kernel Characterization System (SKCS 4100, Perten Instruments, Springfield, Ill.). Kernel hardness was measured on all seven Reference Panel lines for growing years 2011-12 and 2012-13. The hardness indices varied between genotypes. The seven lines ranged from 50.5 to 77.6 in 2011-12 and 35.9 to 78.2 in 2012-13. Nair et al. (2010; 2011a) examined a large set of barley lines for variation in kernel hardness and optimized the test to account for hulled versus hull-less lines. Kernel hardness QTLs have been identified, with the largest one on the short arm of 5H accounting for 22% of the variation in SKCS hardness (Beecher et al., 2002). Hardness affects processing, so it is an important trait to measure in food barley. Nair et al. (2011b) found that hard kernels produced a higher pearling yield than soft kernels, but require additional pearling time. Hard kernels have a more densely packed endosperm, and during milling the starch particles remain trapped in the protein matrix resulting in larger particle sizes, whereas soft kernels result in smaller particles (Nair et al., 2011b). We have seen in our own processing attempts that the softer kernels roll well and the harder kernels crack more effectively. Reference panel grain protein, β-glucan, and kernel hardness data from 2012 and 2013 can be found in Table 1.3.

Doughs and batters are key cereal-processing intermediates, and absorption

capacity is a key functional attribute of any flour used for their manufacture. This is not always just water absorption, but for example the absorption of a concentrated sucrose solution in cookie manufacture. Absorption also has considerable leverage over the quality of the finished product: indirectly through the process intermediates, and directly by affecting moisture content and water activity. For example, all breads require flour that makes dough of a dependable and relatively soft consistency, that can be easily molded into the desired shape, at a level of water absorption that allows the dough to be cohesive and elastic without undue stickiness (Ross and Bettge, 2009). This is no less important for barley flour when used for example, in composite barley-wheat breads, tortillas, or pancakes among others. Given the diversity of barley composition and functionality related to genotype and environment, the ability to monitor and/or control absorption is vital in the acceptance of barley flours in food manufacturing.

Cereal flour absorption capacity is related to kernel hardness, non-starch polysaccharide (NSP) content, and protein content and composition. To monitor these polymeric components in wheat the solvent retention capacity (SRC) method was created (reviewed by Kweon et al., 2011: AACC-International Approved Method 56-11.02). SRC is a composite method that uses four "solvents" to create a functionality fingerprint for a flour: water and three aqueous solutions, 50% w/w sucrose, 2% w/w sodium carbonate, and 5% w/w lactic acid. The four SRCs are determined as the percentage weight increase of the flour pellet after it absorbs the solvents, is centrifuged under controlled conditions, and the supernatant decanted to allow

weighing. The basic principal is that compatible solvents can swell polymeric networks. Different solvents emphasize swelling of different polymeric networks because of differences in solvent/polymer compatibility. The underlying physical chemistry is complex and beyond the scope of this article (see Kweon et al., 2011). Water swells all polymers in cereal flours, sucrose preferentially swells prolamins and NSP (designed for arabinoxylans and not β-glucan), carbonate swells starch damaged in the dry milling process, and lactic acid swells glutelins.

We thought that given the wide ranges of hardness and β-glucan content that we have encountered in barley genotypes that the SRC test could be applied and that it would be as valuable in describing the functionality of barley flour as it is for wheat flour. As we are primarily interested in whole- or minimally pearled barley we attempted to apply the SRC method to flours made from these raw materials. A practical issue arose immediately in two of the solvents (sucrose and carbonate) where two of the Reference Panel entries (Willamette Pearl and Full Pint) showed an inability under standard conditions to create a compacted hydrated-flour pellet after centrifugation. Even with extended or higher g centrifugation compaction could not be achieved. The small number of samples precluded any useful correlation analyses: even commonly highly correlated parameters such as water and carbonate SRC (Ross and Bettge, 2009) were only barely significant ($p = 0.04$) with r values of 0.8. Nonetheless, trends were evident. For example in 2012 DZ100289 and Streaker were ranked lowest for both hardness index and water SRC. There is insufficient data to conclude further at this stage. Further experimentation was conducted on the OFOOD

multi-location trial; the same complications arose for the lines with waxy starch, this time including the lactic acid solvent. Work is ongoing to establish the cause and whether a wetting agent may alleviate the issue.

We were also interested in looking at the mineral composition of the barley grain. The USDA website (http://ndb.nal.usda.gov/) has a basic nutritional profile for barley flour and hulled and pearled grain. This information is based on unnamed varieties and we were intrigued to see how the seven lines in the reference panel would compare. Thanks to Dr. Will Austin from the Central Analytic Lab at OSU, we found the mineral compositions of the seven lines to be higher than or equivalent to the USDA standard for all comparable traits for both years (Tables 1.4 and 1.5). It is unclear what the availability of these micronutrients is in the human diet (Frolich, 1990).

The Traber Lab (led by Dr. Maret G Traber) in the Linus Pauling Institute at Oregon State measured the Vitamin E content in the seven varieties for the 2012 harvest. Only five of the eight components of Vitamin E were measured: delta-tocotrienol, gamma-tocotrienol, alpha-tocotrienol, gamma-tocopherol, and alpha-tocopherol. (B-forms do not elute separately from gamma-tocopherol, and delta-tocopherol was below detection.) The total Vitamin E content differed between the varieties (Fig. 1.4). Tocotrienols are the main form of Vitamin E in cereals, and are primarily located in the pericarp and endosperm, whereas tocopherols are found mainly in the embryo (Brinch-Pedersen et al., 2007). Of the tocotrienols, alpha-tocotrienols are the most prominent, with gamma-tocotrienols following.

Beyond Streaker

Streaker provides growers and consumers with an adapted, winter growth habit hull-less barley. However, our longer term goals in the food barley arena are to increase agronomic performance and provide growers and consumers with a range of grain colors, tastes, textures, and processing attributes. Working with a network of international collaborators, we collected germplasm from Europe and Asia in order to broaden our germplasm base. In order to efficiently introgress this exotic germplasm into adapted backgrounds, we collaborated with Dr. Luis Cistué from the Estación Experimental de Aula Dei, CSIC, in Zaragoza, Spain to produce doubled haploids from the exotic x adapted crosses. In the first phase of this collaboration, we produced doubled haploids (using anther culture) from crosses of OSU food germplasm (described in Chutimanitsakun et al., 2013) with selected German winter barleys that have excellent agronomic performance and high levels of winter hardiness. These doubled haploids were grown in Oregon, USA and at multiple locations in Spain, where Dr. Cistué cooperates with Semillas Batlle (http://semillasbatlle.es/en) and colleagues at the Universidad de Lleida, led by Dr. Ignacio Romagosa.

After several years of phenotypic selection for agronomic and quality traits at these locations, selected lines were advanced to the INUDFOOD trial. There are 30 lines in this trial, 13 selected by OSU, 14 selected by Dr. Cistué and colleagues and three check varieties. The experimental germplasm is all doubled haploid and hull-less and includes waxy and non-waxy starch types with moderately high grain β-glucan content (Fig. 1.1). This trial was planted in Fall 2014 at four locations:

Corvallis, OR, USA; Pullman, WA, USA; Lleida, Spain; and Dundee, Scotland. The cooperators in Washington, USA and Scotland are Dr. Kevin Murphy, Washington State University and Dr. Bill Thomas (James Hutton Institute), respectively. This experiment represents the start of an international collaboration directed at the rapid development of diverse food barley germplasm resources.

At the same time the INUDFOOD lines were being developed, other European germplasm was crossed with OSU food germplasm. The resulting lines are all two-row and hull-less. They are currently in the F6 generation in a trial known as the 'EurOregon 2-rows' (Fig. 1.1), which is being grown at multiple locations in the Pacific Northwest of the USA.

The collaboration with Dr. Cistué in doubled haploid production expanded to include the development of a doubled haploid lab at Oregon State University (http://barleyworld.org/doubled-haploid). This on-site capability was then used to accelerate the development of three major classes of food barley germplasm: the OCOLOR project, the UG99 project, and the doubled haploid genomic selection (DHGS) project. Each of these initiatives will be described in the subsequent narrative.

The OCOLOR germplasm was developed with the goal of introgressing alleles for grain color, aroma, and flavor from accessions collected in Tibet and Nepal by Dr. Kazuhiro Sato (Okayama University Research Institute of Bioresources). There is evidence that barley with colored grain has increased levels of anthocyanins, total phenolics, and antioxidants (Kim et al., 2007; Bellido and B, 2009; Abdel-Aal et

al., 2012; Gong et al., 2012). The strategy was to cross the Himalayan accessions with locally adapted germplasm, advance the progeny through single seed descent (SSD) and select for novel grain types and adaptation to Pacific Northwest, USA conditions in advanced generations (Fig.1.1). After multiple cycles of phenotypic selection, a subset of lines were chosen for accelerated advance to homozygosity via doubled haploid production. These doubled haploids are currently in field trials.

The UG99 project was initiated with the goal of defensively introgressing alleles conferring resistance to the stem rust pathogen (*Puccinia graminis*) race TTKSK, isolate UG99 into our food barley germplasm. As sources of resistance, we targeted the alleles identified by Brueggeman et al. (2009) at the *rpg4/Rpg5* complex located on chromosome 5H. We initiated this process thanks to the generous gift of six accessions from Dr. Aaron Beattie (University of Saskatchewan, Saskatoon, Canada), each of which carried the target resistance alleles. Fortuitously, these accessions included hull-less types. The resistance donors were crossed with a range of locally adapted varieties and germplasm (Fig. 1.1). Selected doubled haploids with hull-less seed will be advanced to field trials for agronomic and quality assessment. Via MAS, lines with target *rpg4/Rpg5* alleles can be selected and advanced to disease resistance confirmation.

The INUDFOOD, OCOLOR, and UG99 projects all represent stand-alone, trait-based plant breeding efforts focused on food barley germplasm enhancement. To date, all our efforts have involved MAS, phenotypic selection, or a combination of both. We have now implemented an integrated genomic selection breeding scheme to

develop the next generation of food barley germplasm. Traditional marker assisted selection, while useful for simply inherited traits controlled by few loci, loses effectiveness as the number of loci increases. This is true for individual quantitative traits or when multiple traits are under selection. Genomic selection uses a training population that has been phenotyped and genotyped to estimate effects for a large set of markers distributed across the genome (Meuwissen et al., 2001). The marker effects are applied to an individual that has only been genotyped to estimate its breeding value (GEBV). The primary benefit of genomic selection is that parents with superior breeding value for quantitative traits can be identified very early in the breeding process substantially reducing the breeding cycle time (Heffner et al., 2010). This allows for an accelerated recurrent selection program. In addition to rapid cycle selection of parents, genomic selection can be applied to segregating inbred or doubled haploid lines derived from early generation parents to predict line performance *per se*.

Promising assessment of genomic selection in animal systems has prompted a flurry of activity exploring the feasibility of genomic selection in plant breeding. Initial optimism was supported by simulation studies that demonstrated greater response to selection using genomic selection compared to conventional marker assisted selection or phenotypic selection (Bernardo, 2008; Iwata and Jannink, 2011). These were followed by empirical studies using cross-validation that further supported advantages of genomic selection (Lorenzana and Bernardo, 2009; Heffner et al., 2010; 2011). Subsequent studies have shown good prediction accuracy can be

obtained with relatively small training populations (hundreds) compared to animals systems that use thousands of individuals (Heffner et al., 2011; Lorenz et al., 2012). Similarly, no significant increase in accuracy occurred when the number of markers increased beyond 384 in barley (Lorenz et al., 2012). In the same study, two closely related breeding programs were used as training and validation sets. Prediction accuracy was greater when the same program was used for the training population and selection candidates indicating that the composition of the training population is an important determinant of prediction accuracy. Comparison of various models to estimate marker effects have generally shown little difference among models and that the model with the simplest assumptions (ridge regression BLUP) can be used effectively (Lorenzana and Bernardo, 2009; Heslot et al., 2012; Zhong et al., 2009; Crossa et al., 2010; Lorenz et al., 2012). Taken together, these studies indicate that using ridge regression BLUP, a training population of 300 individuals that is closely related to the selection candidates, and ~400 markers will be the best approach to generate prediction accuracies that will substantially improve genetic gain per year in a facultative food barley breeding program.

Doubled haploid methods accelerate generation time by creating completely inbred lines from gametes sampled at any generation. In the most common application, F2 gametes are sampled from F1 plants and the resulting array of inbred lines are used for genetic mapping and breeding. In terms of the former, many bi-parental QTL mapping populations have been used effectively in barley and our research groups have been participants in many of these endeavors (reviewed by

Cistué et al., 2011). Most recently, we used doubled haploid populations to identify a new QTL associated with low temperature tolerance (Fisk et al., 2013). In terms of breeding applications, doubled haploids are used extensively in maize for inbred development (Murovec and Bohanec, 2011) and in cereals for variety development (Cistué et al., 2011; Zheng et al., 2002). In barley, there are both gynogenetic (*Hordeum bulbosum*) and androgenetic (anther/microspore culture) available. Our lab has experience with both (Cistué et al., 2011) and within the past year we have implemented anther culture, produced over thousands of DH lines, and offered the service to the research community on a cost-recovery basis (http://barleyworld.org/doubled-haploid). Protocols have developed to the point that genotype specificity is not an issue. Doubled haploid approaches are not a universal solution to plant breeding challenges: Li et al. (2013) reported that conventional advance via shuttle breeding was more advantageous than doubled haploid for the CIMMYT wheat program. While some have expressed doubt in the value and/or efficiency of doubled haploids compared to conventional line development, we see tremendous opportunity in the context of genomic selection, as described in the next section. Furthermore, doubled haploid genetic stocks provide an "immortal" resource for continual re-analysis, improvement, and launching new initiatives.

The food barley breeding program will be developed from three different germplasm pools (Fig. 1.1). Briefly, the composition of the three germplasm pools is as follows: 1) European, Asian and US varieties and breeding lines with different food quality attributes: hulled/hull-les, colored/non-colored and waxy/non-waxy

starch. This germplasm includes the components of Streaker, as well as selections from the OFOOD, INUDFOOD, and the OCOLOR projects, selected varieties from the Cereal Breeding Research Darzau program led by Dr. Karl-Josef Mueller, and food barley germplasm developed by the USDA-ARS program at Aberdeen, Idaho, USA under the direction of Dr. Gongshe Hu; 2) USA varieties and breeding lines with exceptional low temperature tolerance (LTT) (two and six rowed). This germplasm was (i) developed and characterized for the Barley CAP project or (ii) assembled for an extensive LTT association mapping project supported by the Triticeae CAP; and 3) Varieties and breeding lines adapted to the environments that this project will focus on in the USA: Oregon, Washington, Idaho and Minnesota. This germplasm was contributed by the four participating breeding programs and includes sources of resistance to diseases endemic to one or more of the target environments.

The crossing block was designed so that segregation for the traits of interest was maximized i.e., the two parents of each cross usually belong to different germplasm pools and have different attributes (e.g. hulled x hull-less, winter x spring or facultative, colored x non-colored). The training population (C0) will consist of doubled haploids derived from F1s of these crosses and from intermated F1s of these crosses. Remnant seed of the F1xF1 crosses will be used as segregating material in the first cycle of genomic selection. Thereafter, selected lines will be intermated. Through the cycles of genomic selection the frequency of favorable alleles for the traits that are under selection will increase and it is likely that some alleles may

become fixed. The flexibility of our genotyping approach allows for updating marker panels to maximize genetic information at each cycle. Since the training population will be extensively characterized phenotypically and genotypically there is always the option to go back to the original F1s and create a new starting population based on completely different traits that may be of interest in the future. The prediction model is also continuously rejuvenated as genotypic and phenotypic information from elite lines derived from the participating breeding programs is incorporated into the prediction models. In this way, new germplasm can be infused into the system at any point. As lines derived from the newly infused germplasm advance in the breeding process, their genotypic and phenotypic information can also be incorporated into the prediction models. (Process described in Fig. 1.5).

The breeding goals of this project are varieties with strong agronomic performance, abiotic/biotic resistances, and a range of quality traits suited to different food end-uses. Our baseline check is Streaker. For agronomic and resistance traits, the goals are clear: higher yield and low temperature tolerance than the check. Agronomic variables contributing to yield are lodging resistance and grain test weight. For food quality, there are opportunities to develop a range of products, each with contrasting quality attributes. The baseline quality criterion is grain β-glucan higher than Streaker. Beyond that we will maintain genetic/phenotypic diversity for the other quality attributes with the framework of an overall goal of four principal germplasm types (all hull-less) that represent all possible combinations of starch type (waxy: non-waxy) and grain color (white: colored).

We propose through the doubled haploid material to characterize quality and create a naming system that could be proposed to the grower and processor communities as a model for commercial classification of food barley. Within the 300 doubled haploids in the training population we anticipate finding most combinations of these categorizable factors.

Conclusion

Despite an overall decline in food barley consumption for the last few centuries, food barley germplasm development and quality characterization is making a comeback in many areas of the world. As consumers realize the nutrition and taste benefits of barley, commercial production increases and there becomes a need for new varieties adapted to a number of different regions. Oregon State University is on the forefront of food barley research, thanks to extensive national and international collaboration. With new and exciting germplasm just down the pipeline, we hope to invigorate our local barley market by engaging farmers, processors, and consumers and to assist in developing markets worldwide.

Acknowledgements

This research has been made possible by the dedicated efforts of OSU Barley Project members (current and past), collaborators around the world, and funding provided by visionary sponsors. Special thanks to Tanya Filichkin, for her critical roles in doubled haploid production, germplasm advance, and genotyping and to Ann

Corey for her assistance with germplasm development and assessment. Thanks to Teepakorn Kongraksawech for his assistance with quality assessments. Thanks to Emily Watts in the Traber Lab for running the Vitamin E tests. Thanks to Pierrette Castro for her assistance with the manuscript references. This research was supported by the Oregon State University Agricultural Experiment Station, Formula Grant no. 2013-31100-06041 from the United States Department of Agriculture National Institute of Food and Agriculture, the Triticeae Coordinated Agricultural Project Grant no. 2011-68002-30029 from the United States Department of Agriculture, National Institute of Food and Agriculture, the Oregon State University Research Foundation, The Oregon Wheat Commission, and the Idaho Barley Commission.

References

AACC International. Approved Methods of Analysis, 11th Ed. Method 56-11.02. Solvent Retention Capacity Profile. Approved June 3, 2009.AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-56-11.02

AACC International. Approved Methods of Analysis, 11th Ed. Method 32-23.01. β-glucan Content of Barley and Oats- Rapid Enzymatic Procedure. Approved November 3, 1999. AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-32-23.01

Abdel-Aal ESM, Choo TM, Dhillon S, Rabalski I (2012) Free and bound phenolic acids and total phenolics in black, blue, and yellow barley and their contribution to free radical scavenging capacity. Cereal Chem. 89(4): 198-204

Abraha A, Uhlen AK, Abay F, Sahlstrøm S, Bjørnstad Å (2013) Genetic variation in barley enables a high quality, the Ethiopian staple flat bread, comparable to Tef. Crop Sci. 53(5): 2040-2050

AbuMweis SS, Jew S, Ames NP (2010) β-glucan from barley and its lipid-lowering capacity: a meta-analysis of randomized, controlled trials. Eur J Clin Nutr 64: 1472-1480

Ames NP and Rhymer CR (2008) Issues surrounding health claims for barley. J Nutr 138: 1237S-1243S

Ames N, Rhymer C, Rossnagel B, Therrien M, Ryland D, Dua S, Ross K (2006) Utilization of diverse hulless barley properties to maximize food product quality. Cereal Food World. 51(1): 23-28

Arndt EA (2006) Whole-grain barley for today's health and wellness needs. Cereal Food World. 51: 20-22

Baik BK and Czuchajowska Z (1997) Barley in udon noodles. Food Sci Technol Int. 3: 423-435

Baik BK, Newman CW, Newman RK (2011) Food uses of barley. In: Ullrich SE (eds) Barley: Production, Improvement, and Uses. Wiley-Blackwell, 532-562

Baik BK and Ullrich SE (2008) Barley for food: characteristics, improvement, and renewed interest. J Cereal Sci. 48: 233-242

Bays H, Frestedt JL, Bell M, Williams C, Kolberg L, Schmelzer W, Anderson JW (2011) Reduced viscosity barley β-glucan versus placebo: a randomized controlled trial of the effects on insulin sensitivity for individuals at risk for diabetes mellitus. Nutr Metab. 8: 10

Beecher B, Bowman J, Martin JM, Bettge AD, Morris CF, Blake TK, Giroux MJ (2002) Hordoindolines are associated with a major endosperm- texture QTL in barley (Hordeum vulgare). Genome. 45(3): 584-591

Behall KM, Scholfield DJ, Hallfrisch JG (2006) Barley β-glucan reduces plasma glucose and insulin responses compared with resistant starch in men. Nutr Res. 26: 644-650

Behall KM, Scholfield DJ, Hallfrisch JG (2004a) Diets containing barley significantly reduce lipids in mildly hypercholesterolemic men and women. Am J Clin Nutr. 80(5): 1185-1193

Behall KM, Scholfield DJ, Hallfrisch JG (2004b) Lipids significantly reduced by diets containing barley in moderately hypercholesterolemic men. J Am Coll Nutr. 23: 55-62

Bellido GG and B T (2009) Anthocyanin composition and oxygen radical scavenging capacity (ORAC) of milled and pearled purple, black, and common barley. J Agr Food Chem. 57(3): 1022-1028

Bendelow VM (1975) Determination of non-starch polysaccharides in barley breeding programmes. J I Brewing. 81(2): 127-130

Bernardo R (2008) Molecular markers and selection for complex traits in plants: learning from the last 20 years. Crop Sci. 48: 1649–1664

Biorklund M, van Rees A, Mensink RP, Onning G (2005) Changes in serum lipids and postprandial glucose and insulin concentrations after consumption of beverages with β-glucans from oats or barley: a randomised dose-controlled trial. Eur J Clin Nutr. 59: 1272–1281

Brennan CS and Cleary LJ (2005) The potential use of cereal (1→ 3, 1→ 4)-β-D-glucans as functional food ingredients. J Cereal Sci. 42(1): 1-13

Brinch-Pedersen H, Borg S, Tauris B, Holm PB (2007) Molecular genetic approaches to increasing mineral availability and vitamin content of cereals. J Cereal Sci. 46(3): 308-326

Brooks WS, Griffey CA, Price AM, Pridgen TH, Rohrer WL (2005) Registration of 'Doyce' Hulless Barley the ventral furrow. Crop Sci. 45: 791-792

Brouns F, Kettlitz B, Arrigoni E (2002) Resistant starch and "the butyrate revolution". Trends Food Sci Tech. 13(8): 251-261

Brueggeman R, Steffenson BJ, Kleinhofs A (2009) The rpg4/Rpg5 stem rust resistance locus in barley. Cell Cycle. 8(7): 977-981

Cascadia Grains Conference 2013 http://cascadiagrains.com/events-registration/past-events/2013-conference/ accessed 2014-02-12

Cascadia Grains Conference 2014 http://cascadiagrains.com/events-registration/past-events/2014-conference/ accessed 2014-02-12

Casiraghi MC, Garsetti M, Testolin G, Brighenti F (2006) Post-prandial responses to cereal products enriched with barley β-glucan. J Am Coll Nutr. 25: 313-320

Champ M (2013) Metabolic Effects of Resistant Starch. In: Shi YC and Maningat CC (eds.) Resistant Starch: Sources, Applications and Health Benefits. John Wiley & Sons, 2013 p. 229-249

Cho SS, Prosky L, Dreher M (1999) Complex carbohydrates in food. Marcel Dekker, New York.

Chutimanitsakun Y, Cuesta-Marcos A, Chao S, Corey A, Filichkin T, Fisk S, Kolding M, Meints B, Ong YL, Rey JI, Ross AS, Hayes PM (2013) Application of marker-assisted selection and genome-wide association scanning to the development of winter food barley germplasm resources. Plant Breeding. 132(6): 563-570

Cistué L, Cuesta-Marcos A, Chao S, Echávarri B, Chutimanitsakun Y, Corey A, Filichkin T, Garcia-Mariño N, Romagosa I, Hayes P (2011) Comparative mapping of the Oregon Wolfe Barley using doubled haploid lines derived from female and male gametes. Theor Appl Genet. 122: 1399-1410

Crosbie GB and Ross AS (2004) Asian wheat flour noodles. In: Wrigley CW, Corke H, Walker CE (eds.) The Encyclopedia of Grain Science, Vol 2. Elsevier Academic Press, Oxford UK, 304-312

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

Dai F, Nevo E, Wu D, Comadran J, Zhou M, Qiu L, Chen Z, Beiles A, Chen G, Zhang G (2012) Tibet is one of the centers of domestication of cultivated barley. P Natl Acad Sci USA. 109(42): 16969-16973

Di Cagno R, De Angelis M, Lavermicocca P, De Vincenzi M, Giovannini C, Faccia M, Gobbetti M (2002) Proteolysis by sourdough lactic acid bacteria: effects on wheat flour protein fractions and gliadin peptides involved in human cereal intolerance. Appl Environ Microb. 68(2): 623-633

Edney MJ, Rossnagel BG, Endo Y, Ozawa S, Brophy M (2002) Pearling quality of Canadian barley varieties and their potential use as rice extenders. J Cereal Sci. 36: 295-305

FAOSTAT (2011) Production and trade. (http://faostat.fao.org/site/339/default.aspx) Verified February 13, 2013. FAO, Rome, Italy

Fardet A (2010) New hypotheses for the health-protective mechanisms of whole-grain cereals: what is beyond fibre? Nutr Res Rev. 23.01: 65-134

Fardet A (2013) Whole Grains from a Mechanistic View. Proceedings of the Whole Grains Summit 2012. T. Schryver Ed. http://www.aaccnet.org/publications/plexus/cfwplexus/library/books/Documents/WholeGrainsSummit2012/CPLEX-2013-1001-01B.pdf accessed 2014-02-14

Fardet A and Rock E (2013) The search for a new paradigm to study micronutrient and phytochemical bioavailability: From reductionism to holism. Med Hypotheses.http://dx.doi.org/10.1016/j.mehy.2013.11.035

Fastnaught CE (2010) Barley fiber. In: Cho SS and Samuel P (eds.) Fiber Ingredients: Food Applications and Health Benefits. CRC Press, 323-358

Fisk SP, Cuesta-Marcos A, Cistue L, Russell J, Smith K, Baenziger S, Bedo Z, Corey A, Filichkin T, Karsai I, Waugh R, Hayes PM (2013) FR-H3: a new QTL to assist in the development of fall-sown barley with superior low temperature tolerance. Theor Appl Genet. 126: 335-347

Frølich W (1990) Chelating properties of dietary fiber and phytate. The role for mineral availability. In: New Developments in Dietary Fiber. Springer US, 83-93

Fung KY, Cosgrove L, Lockett T, Head R, Topping DL (2012) A review of the potential mechanisms for the lowering of colorectal oncogenesis by butyrate. Brit J Nutr. 1(1): 1-1

Gänzle MG, Loponen J, Gobbetti M (2008) Proteolysis in sourdough fermentations: mechanisms and potential for improved bread quality. Trends Food Sci Tech. 19(10): 513-521

Gong LX, Jin C, Wu LJ, Wu XQ, Zhang Y (2012) Tiβn Hull-less Barley (Hordeum vulgare L.) as a potential source of antioxidants. Cereal Chem. 89(6): 290-295

Grando S and Macpherson HG (2005) (eds.) Food Barley: Importance, Uses and Local Knowledge. Proceedings of the International Workshop on Food Barley Improvement, 14-17 January 2002, Hammamet, Tunisia. ICARDA, Aleppo, Syria, x+156 pp. En.

Gray D, Abdel-Aal ESM, Seetharaman K, Kakuda Y (2010) Differences in viscosity and textural properties of selected barley cultivars as influenced by pearling and cooking. Food Chem. 120: 402-409

Gupta M, Bawa AS, Semwal AD (2008) Effect of barley flour incorporation on the instrumental texture of sponge cake. Int J Food Pro. 12: 243-251

Hatcher DW, Lagasse S, Dexter JE, Rossnagel B, Izydorczyk M (2005) Quality characteristics of yellow alkaline noodles enriched with hull-less barley flour. Cereal Chem. 82: 60-69

Heffner E, Jannink JL, Sorrells ME (2011) Genomic selection accuracy using multifamily prediction models in a wheat breeding program. Plant Gen. 4: 65–75

Heffner EL, Lorenz AJ, Jannink JL, Sorrells ME (2010) Plant breeding with genomic selection: gain per unit time and cost. Crop Sci. 50: 1681–1690

Heslot N, Yang HP, Sorrells ME, Jannink JL (2012) Genomic selection in plant breeding: a comparison of models. Crop Sci. 52: 146-160

Hinata M, Ono M, Midorikawa S, Nakanishi K (2007) Metabolic improvement of male prisoners with type 2 diabetes in Fukushima Prison, Japan. Diabetes Res Clin Pr. 77(2): 327-332

Holtekjolen AK, Sahlstrom S, Knutsen SH (2011) Phenolic contents and antioxidant activities in covered whole-grain flours of Norwegian barley varieties and in fractions obtained after pearling. Acta Agric Scand. 61: 67-74

Hu G and Burton C (2008) Modification of standard enzymatic protocol to a cost-efficient format for mixed-linkage (1→3,1→4)-β-d-glucan measurement. Cereal Chem. 85: 648-65

Islamovic E, Obert DE, Oliver RE, Harrison SA, Ibrahim A, Marshall JM, Miclaus KJ, Hu G, Jackson EW (2013) Genetic dissection of grain β-glucan and amylose content in barley (*Hordeum vulgare* L.). Mol Breeding. 31: 15-25

Iwata H and Jannink JL (2011) Accuracy of genomic selection prediction in barley breeding programs: A simulation study based on the real single nucleotide polymorphism data of barley breeding lines. Crop Sci. 51: 1915–1927

Izydorczyk MS (2010) Variations in content and molecular structure of barley nonstarch polysaccharides associated with genotypic and cellular origin. Cereal Chem. 87: 376-384

Izydorczyk MS, Chornick TL, Paulley FG, Edwards NM, Dexter JE (2008) Physicochemical properties of hull-less barley fibre-rich fractions varying in particle size and their potential as functional ingredients in two-layer flat bread. Food Chem. 108: 561-570

Izydorczyk MS and Dexter JE (2008) Barley β-glucans and arabinoxylans: molecular structure, physicochemical properties, and uses in food products–a review. Food Res Int. 41(9): 850-868

Izydorczyk MS, Lagasse S, Hatcher DW, Dexter JE, Rossnagel BG (2005) The enrichment of Asian noodles with fiber-rich fractions derived from roller milling of hull-less barley. J Sci Food Agr. 85: 2094-2104

Jones JM (2010) Dietary Fibre‘s Co-Passengers: Is It the Fibre or the Co-Passengers pp 365-378. In:. Van Der Kamp J.W., Jones J, Mccleary B, Topping DJ (eds.) Dietary Fibre: New Frontiers for Food and Health. Wageningen Academic Publishers: Wageningen, The Netherlands

Jones JM, Reicks M, Adams J, Fulcher G, Weaver G, Kanter M, Marquart L (2002) The importance of promoting a whole grain foods message. J Am Coll Nutr. 21(4): 293-297

Keenan JM, Goulson M, Shamliyan T, Knutson N, Kolberg L, Curry L (2007) The effects of concentrated barley β-glucan on blood lipids in a population of hypercholesterolaemic men and women. Brit J Nutr. 97(06): 1162-1168

Keogh GF, Cooper GJ, Mulvey TB, McArdle BH, Coles GD, Monro JA, Poppitt SD (2003) Randomized controlled crossover study of the effect of a highly β-glucan–enriched barley on cardiovascular disease risk factors in mildly hypercholesterolemic men. Am J Clin Nutr. 78(4): 711-718

Kim H, Stote KS, Behall KM, Spears K, Vinyard B, Conway JM (2009) Glucose and insulin responses to whole grain breakfasts varying in soluble fiber, β-glucan. Eur J Nutr. 48: 170-175

Kim H, Turowski M, Anderson WK, Young SA, Kim Y, Yokoyama W (2011a) Supplementation of hydroxypropyl methylcellulose into yeast leavened all-whole grain barley bread potentiates cholesterol-lowering effect. J Agr Food Chem. 59(14): 7672-7678

Kim MJ, Hyun JN, Kim JA, Park JC, Kim MY, Kim JG, Lee SJ, Chun SC, Chung IM (2007) Relationship between phenolic compounds, anthocyanins content and antioxidant activity in colored barley germplasm. J Agr Food Chem. 55(12): 4802-4809

Kim Y and Yokoyama WH (2011b) Physical and sensory properties of all-barley and all-oat breads with additional Hydroxypropyl Methylcellulose (HPMC) β-glucan. J Agr Food Chem. 59: 741-746

King RA, Noakes M, Bird AR, Morell MK, Topping DL (2008) An extruded breakfast cereal made from a high amylose barley cultivar has a low glycemic index and lower plasma insulin response than one made from a standard barley. J Cereal Sci. 48: 526-530

Kinner M, Nitschko S, Sommeregger J, Petrasch A, Linsberger-Martin G, Grausgruber H, Berghofer E, Siebenhandl-Ehn S (2011) Naked barley—optimized recipe for pure barley bread with sufficient β-glucan according to the EFSA health claims. J Cereal Sci. 53: 225-230

Kweon M, Slade L, Levine H (2011) Solvent Retention Capacity (SRC) Testing of Wheat Flour: Principles and value in predicting flour functionality in different wheat-based food processes and in wheat breeding-A review. Cereal Chem. 88(6): 537-552

Lagassé SL, Hatcher DW, Dexter JE, Rossnagel BG, Izydorczyk MS (2006) Quality characteristics of fresh and dried white salted noodles enriched with flour from hull-less barley genotypes of diverse amylose content. Cereal Chem. 83: 202-210

Li H, Singh RP, Braun HJ, Pfeiffer WH, Wang J (2013) Doubled haploids versus conventional breeding in CIMMYT wheat breeding programs. Crop Sci. 52: 74–83

Li J, Kaneko T, Qin LQ, Wang J, Wang Y (2003) Effects of barley intake on glucose tolerance, lipid metabolism, and bowel function in women. Nutrition. 19(11): 926-929

Lorenz AJ, Smith KP, Jannink JL (2012) Potential and optimization of genomic selection for Fusarium head blight resistance in six-row barley. Crop Sci. 52: 1609-1621

Lorenzana RE and Bernardo R (2009) Accuracy of genotypic value predictions for marker-based selection in bi-parental plant populations. Theor Appl Genet. 120: 151–161

Meuwissen TH, Hayes BJ, Goddard ME (2001) Prediction of total genetic value using genome-wide dense marker maps. Genetics. 157: 1819–1829

Morrell PL and Clegg MT (2007) Genetic evidence for a second domestication of barley (Hordeum vulgare) east of the Fertile Crescent. P Natl Acad Sci USA. 104(9): 3289-3294

Murovec J and Bohanec B (2011) Haploids and doubled haploids in plant breeding. In: Plant breeding. I. Abdurakhmonov (ed.) InTech

Murphy K, Lammer D, Lyon S, Carter B, Jones SS (2005) Breeding for organic and low-input farming systems: An evolutionary–participatory breeding method for inbred cereal grains. Renew Agr Food Syst. 20(01): 48-55

Nair S, Ullrich SE, Blake TK, Cooper B, Griffey CA, Hayes PM, Hole DJ, Horsley RD, Obert DE, Smith KP, Muehlbauer GJ, Baik, B-K (2010) Variation in kernel hardness and associated traits in US barley breeding lines. Cereal Chem. 87: 461-466

Nair S, Knoblauch M, Ullrich S, Baik BK (2011a) Microstructure of hard and soft kernels of barley. J Cereal Sci. 54(3): 354-362

Nair S, Ullrich SE, Baik BK (2011b) Association of barley kernel hardness with physical grain traits and food processing parameters. Cereal Chem. 88(2): 147-152

National Barley Foods Council (2003) Petition for unqualified health claim: Barley β-glucan soluble fiber and barley products containing β-glucan soluble fiber and coronary heart disease. Submitted to the FDA on September 25, 2003.

Nevo E and Shewry PR (1992) Origin, evolution, population genetics and resources for breeding of wild barley, Hordeum spontaneum, in the Fertile Crescent. Barley: genetics, biochemistry, molecular biology and biotechnology. 19-43

Newman RK and Newman CW (2008) Barley for food and health: Science, technology, and products. John Wiley & Sons.

Newton AC, Flavell AJ, George TS, Leat P, Mullholland B, Ramsay L, Bingham IJ (2011) Crops that feed the world 4. Barley: a resilient crop? Strengths and weaknesses in the context of food security. Food Security. 3(2): 141-178

Nugent AP (2005) Health properties of resistant starch. British Nutrition Foundation, Nutrition Bulletin. 30: 27–54

Park Y, Subar AF, Hollenbeck A, Schatzkin A (2011) Dietary fiber intake and mortality in the NIH-AARP diet and health study. Arch Intern Med. 171: 1061-1068

Patron NJ, Smith AM, Fahy BF, Hylton CM, Naldrett MJ, Rossnagel BG, Denyer K (2002) The altered pattern of amylose accumulation in the endosperm of low-amylose barley cultivars is attributable to a single mutant allele of granule-bound starch synthase I with a deletion in the 5′-non-coding region. Plant Physiol. 130(1): 190-198

Prasopsunwattana N, Omary MB, Arndt EA, Cooke PH, Flores RA, Yokoyama W, Toma A, Chongcham S, Lee SP (2009) Particle size effects on the quality of flour tortillas enriched with whole grain waxy barley. Cereal Chem. 86: 439-451

PubMed Health (2011) U.S> National Library of Medicine. (http://www.ncbi.nlm.nih.gov/pubmedhealth/PMH0004546/) Verified February 13, 2013.

Quinde Z, Ullrich SE, Baik BK (2004) Genotypic variation in color and discoloration potential of barley-based food products. Cereal Chem. 81: 752-758

Quinde-Axtell Z, Ullrich SE, Baik BK (2005) Genotypic and environmental effects on color and discoloration potential of barley in food products. Cereal Chem. 82: 711-716

Rey JI, Hayes PM, Petrie SE, Corey A, Flowers M, Ohm JB, Ong C, Rhinhart K, Ross AS (2009) Production of dryland barley for human food: Quality and agronomic performance. Crop Sci. 49(1): 347-355

Rieder A, Holtekjølen AK, Sahlstrøm S, Moldestad A (2012) Effect of barley and oat flour types and sourdoughs on dough rheology and bread quality of composite wheat bread. J Cereal Sci. 55(1): 44-52

Risgaard H (2012) Perler fra Skærtoft Mølle. Politikens Forlag, København, Danmark

Robertson JA, Majsak-Newman G, Ring SG, Selvendran RR (1997) Solubilisation of mixed linkage (1→ 3),(1→ 4) β-D-glucans from barley: effects of cooking and digestion. J Cereal Sci. 25(3): 275-283

Ross AS (2011) (http://blogs.oregonstate.edu/deliciousnessw09/2011/09/29/more-kneading-conference-west-photos/) Verified February 13, 2013.

Ross AS (2013) Whole-grain and fiber-enriched noodles. In: Delcour J.A. and Poutanen K. (eds.) Wholegrain and fiber enriched foods: improving quality. Woodhead Publishing. In Press.

Ross AS and Bettge AD (2009) Passing the test on wheat end-use quality. In: Carver B. (ed.) Wheat: science and trade. Wiley-Blackwell Publishing, pp 455-493

Salamini F, Özkan H, Brandolini A, Schäfer-Pregl R, Martin W (2002) Genetics and geography of wild cereal domestication in the Near East. Nat Rev Genet. 3(6): 429-441

Savin R, Stone PJ, Nicolas ME, Wardlaw IF (1997) Effects of heat stress and moderately high temperature on grain growth and malting quality of barley. Aust J Agr Res. 48: 615-624

Shimizu C, Kihara M, Aoe S, Araki S, Ito K, Hayashi K, Watari J, Sakata Y, Ikegami S (2008) Effect of high β-glucan barley on serum cholesterol concentrations and visceral fat area in Japanese men - A randomized, double-blinded, placebo-controlled trial. Plant Food Hum Nutr. 63: 21-25

Skendi A, Biliaderis CG, Papageorgiou M, Izydorczyk MS (2010) Effects of two barley β-glucan isolates on wheat flour dough and bread properties. Food Chem. 119: 1159-1167

Slavin JL (2005) Dietary fiber and body weight. Nutrition. 21: 411-418

Slavin J, Tucker M, Harriman C, Jonnalagadda SS (2013) Whole Grains: Definition, Dietary Recommendations, and Health Benefits. Cereal Food World. 58(4): 191-198

Sullivan P, Arendt E, Gallagher E (2013) The increasing use of barley and barley by-products in the production of healthier baked goods. Trends Food Sci Tech. 29(2): 124-134 (http://www.sciencedirect.com/science/article/pii/S0924224412002130)

Svihus B and Gullord M (2002) Effect of chemical content and physical characteristics on nutritional value of wheat, barley and oats for poultry. Anim Feed Sci Tech. 102: 71-92

Szczodrak J, Czuchajowska Z, Pomeranz Y (1992) Characterization and estimation of barley polysaccharides by near-infrared spectroscopy. II: Estimation of total β-D-glucans. Cereal Chem. 69(4): 419-423

Szűcs P, Skinner JS, Karsai I, Cuesta-Marcos A, Haggard KG, Corey AE, Chen TH, Hayes PM (2007) Validation of the VRN-H2/VRN-H1 epistatic model in barley reveals that intron length variation in VRN-H1 may account for a continuum of vernalization sensitivity. Mol Genet Genomics. 277(3): 249-261

Taketa S, Amano S, Tsujino Y, Sato T, Saisho D, Kakeda K, Takeda K (2008) Barley grain with adhering hulls is controlled by an ERF family transcription factor gene regulating a lipid biosynthesis pathway. P Natl Acad Sci USA. 105(10): 4062-4067

Taketa S, Kikuchi S, Awayama T, Yamamoto S, Ichii M, Kawasaki S (2004) Monophyletic origin of naked barley inferred from molecular analyses of a marker closely linked to the naked caryopsis gene (nud). Theor Appl Genet. 108(7): 1236-1242

Thondre PS and Henry CJK (2009) High-molecular-weight barley β-glucan in chapatis (unleavened Indian flatbread) lowers glycemic index. Nutr Res. 29: 480-486

Tiwari U and Cummins E (2011) Meta-analysis of the effect of β-glucan intake on blood cholesterol and glucose levels. Nutrition. 27: 1008-1016

Tiwari U, Cummins E, Sullivan P, Flaherty J, Brunton N, Gallagher E (2011) Probabilistic methodology for assessing changes in the level and molecular weight of barley β-glucan during bread baking. Food Chem. 124(4): 1567-1576

Topping DL and Clifton PM (2001) Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. Physiol Rev. 81(3): 1031-1064

Tosh SM, Brummer Y, Miller SS, Regand A, Defelice C, Duss R, Wolever TMS, Wood PJ (2010) Processing affects the physicochemical properties of β-glucan in oat bran cereal. J Agr Food Chem. 58: 7723-7730

van Amelswoort JMM and Westrate JA (1992) Amylose amylopectin ratio in a meal affects postprandial variables in male volunteers. Am J Clin Nutr. 55: 712-718

Vavilov NI (1951) The origin, variation, immunity and breeding of cultivated plants. Soil Sci. 72(6): 482

Verardo V, Gomez-Caravaca AM, Messia MC, Marconi E, Caboni MF (2011) Development of functional spaghetti enriched in bioactive compounds using barley coarse fraction obtained by air classification. J Agr Food Chem. 59: 9127-9134

Vitaglione P, Lumaga RB, Montagnese C, Messia MC, Marconi E, Scalfi L (2010) Satiating effect of a barley β-glucan-enriched snack. J Am Coll Nutr. 29: 113-121

Wolever TMS, Tosh SM, Gibbs AL, Brand-Miller J, Duncan AM, Hart V, Lamarche B, Thomson BA, Duss R, Wood PJ (2010) Physicochemical properties of oat β-glucan influence its ability to reduce serum LDL cholesterol in humans: A randomized clinical trial. Am J Clin Nutr. 92: 723-732

Wolfe MS, Baresel JP, Desclaux D, Goldringer I, Hoad S, Kovacs G, Löschenberger F, Miedaner T, Østergård H, van Bueren ETL (2008) Developments in breeding cereals for organic agriculture. Euphytica. 163(3): 323-346

Wood PJ, Beer MU, Butler G (2000) Evaluation of role of concentration and molecular weight of oat β-glucan in determining effect of viscosity on plasma glucose and insulin following an oral glucose load. Brit J Nutr. 84(01): 19-23

Wood PJ, Weisz J, Beer MU, Newman CW, Newman RK (2003) Structure of (1→ 3)(1→ 4)-β-d-glucan in waxy and nonwaxy barley. Cereal Chem. 80(3): 329-332

Xue Q, Wang L, Newman RK, Newman CW, Graham H (1997) Influence of the hulless, waxy starch and short-awn genes on the composition of barleys. J Cereal Sci. 26(2): 251-257

Zheng GH, Rossnagel BG, Tyler RT, Bhatty RS (2000) Distribution of β-glucan in the grain of hull-less barley. Cereal Chem. 77(2): 140–144

Zheng MY, Weng Y, Liu W, Konzak CF (2002) The effect of ovary-conditioned medium on microspore embryogenesis in common wheat (*Triticum aestivum* L.). Plant Cell Rep. 20: 802–807

Zhong S, Dekkers JC, Fernando RL, Jannink JL (2009) Factors affecting accuracy from genomic selection in populations derived from multiple inbred lines: a barley case study. Genetics. 182(1): 355-364

Figure 1.1. Description of food barley germplasm used in the Oregon State barley breeding program starting in 2005 with the first crosses contributing to the OFOOD trial to the DHGS program in 2014. This figure details the diverse germplasm that has contributed to the different trials described in this chapter.

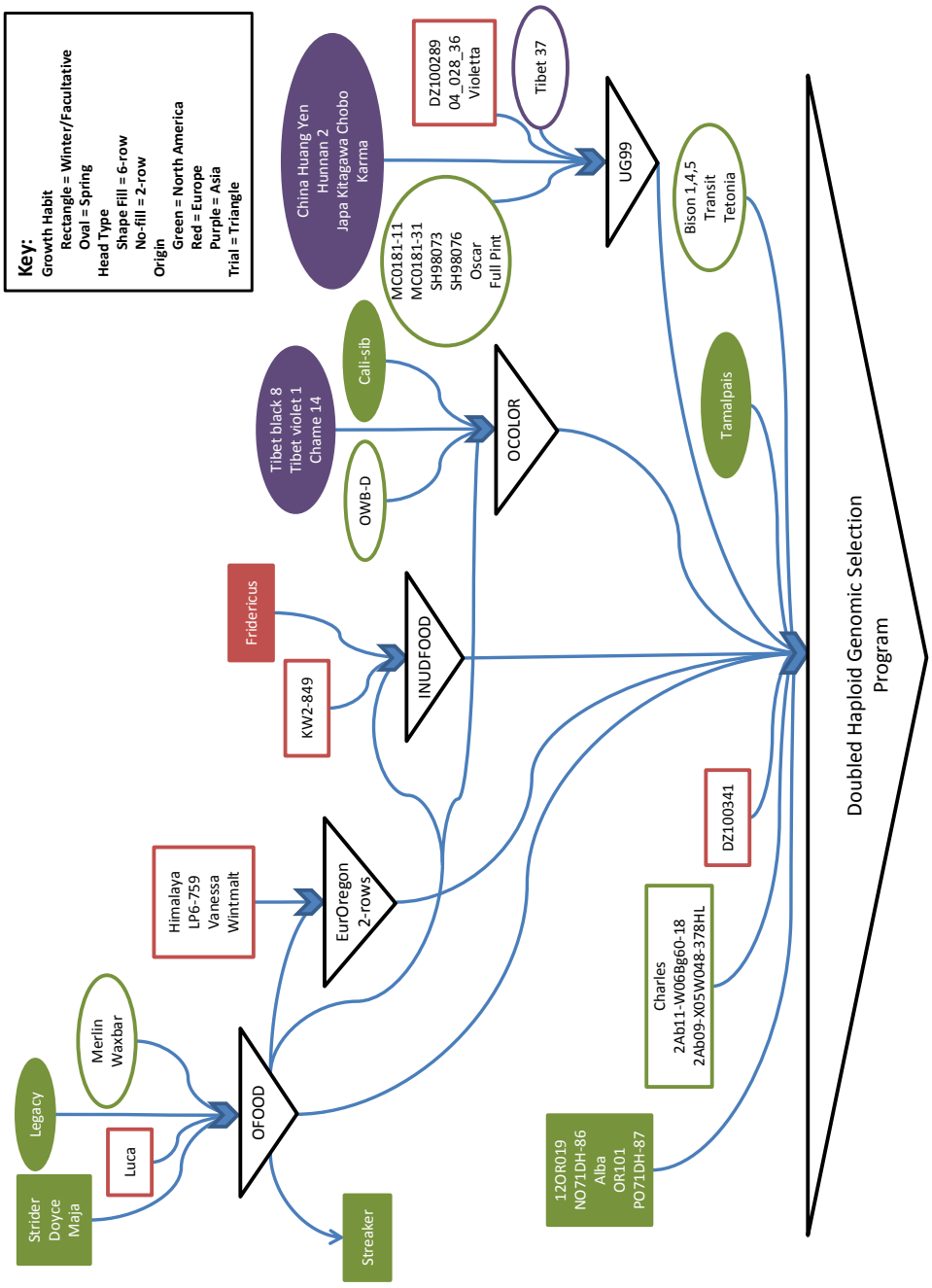


Figure 1.2. Graph showing the mean yield rankings (1 = highest yield) compared to standard deviations of the rank for the OFOOD trial over two years at six locations. Green lines indicate median value.

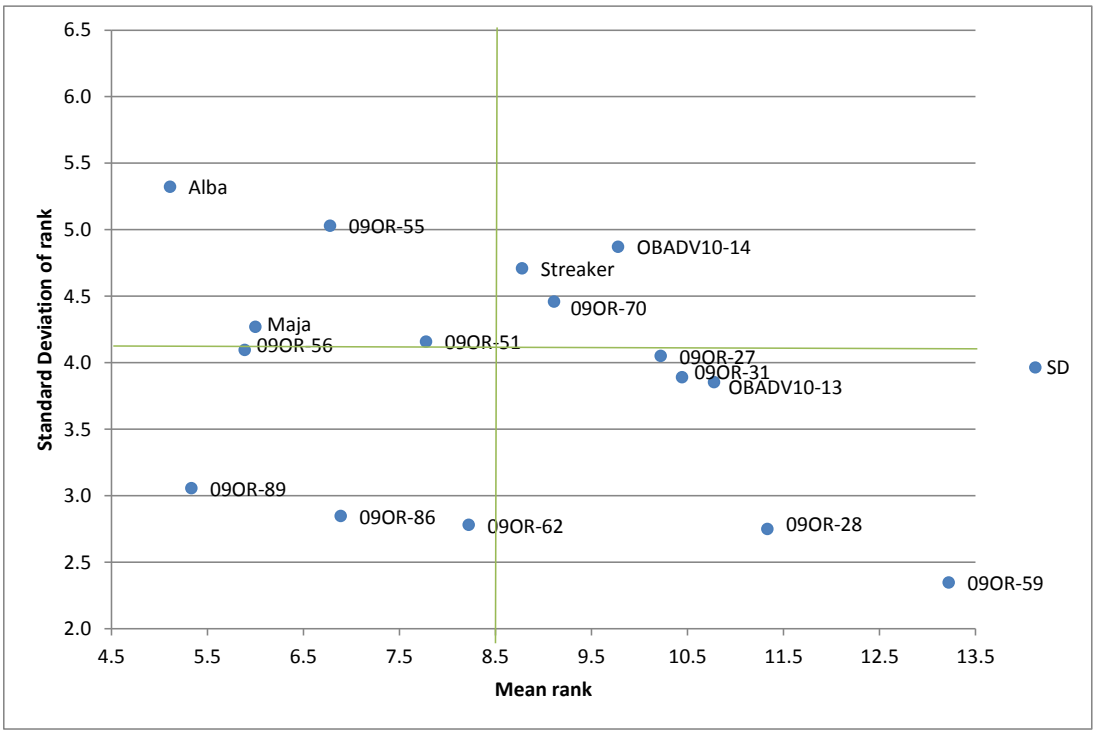


Table 1.1. β-glucan, grain protein, and yield measurements for Streaker at three locations representing different climate conditions in 2011-12 and 2012-13. *β-glucan values for 2011-12 only

	β-glucan (% w/w)*	Grain protein (%)	Yield (kg/ha)
Corvallis, OR (High Rainfall)	4.4	11.8	4635
Aberdeen, ID (Irrigated)	3.6	12.0	8952
Pullman, WA (Dryland)	4.9	13.2	5951
Average across locations	4.3	12.3	6513

Figure 1.3. Streaker Barley Flakes from Camas Country Mill (Alvadore, OR, USA).

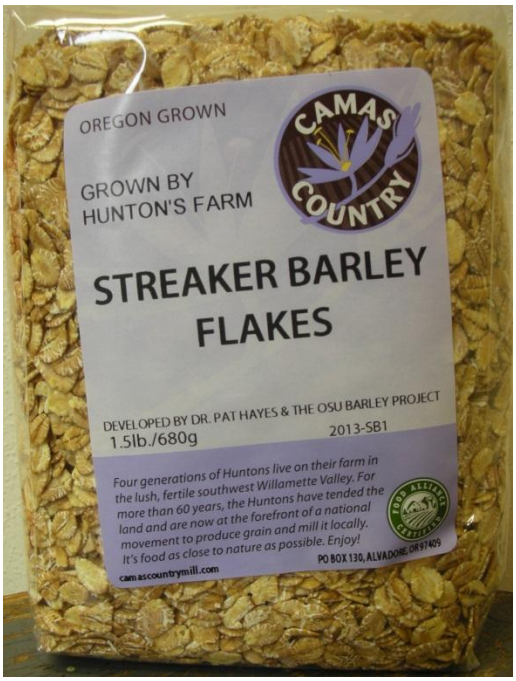


Table 1.2. Barley Breakfast Bar recipe served at the Bethel School District (Lane County, OR, USA). The recipe was adapted to include barley flakes, like those shown in Fig. 3.

Bethel School District

Page 1 — Recipe Sizing Report — Jan 23, 2014

000645 - Bethel RASP Breakfast Bar TEST :	Components	Attributes
HACCP Process: No HACCP Process	Meat/Alt:	
Number of Portions: 200	Grains: 1.25 oz	
Size of Portion: 1 PIECE	Fruit:	
	Vegetable:	
	Milk:	

Ingredients	Measures	Instructions
903062 Rolled Barley Flakes	1 1/2 gals	Combine flour, barley flakes, brown sugar and baking soda and mix well.
020081 FLOUR,ALL PURPOSE WHITE,ENRICHED,BLEACHE...	1 1/2 gals	Reserve 8 cups (2 cups per pan) of this blend. Add canola oil and Applesauce to the dry ingredients.
018372 BAKING SODA	2 Tbsp	
019334 SUGAR,BROWN	1 GAL (unpacked)	
009020 APPLESAUCE,CANNED,SWTND,WO/SALT	1 qt	
004582 OIL,CANOLA	1 qt + 3 cups	
		Add canola oil and applesauce to dry ingredients.
105941 RASPBERRY FRUIT TOPPING	3 qts	Divide mixture in four. Press into an ungreased sheet pan. Spread with raspberry filling/preserve. Sprinkle with reserve mixture - 2 cups per pan.
		Bake at 300 for 30-35 minutes.
		(Cut each pan 10 X 5 to be done at schools)
		This recipe makes 4 x 50 ea pans)

*Nutrients are based upon 1 Portion Size (1 PIECE)

Calories	261	kcal	Cholesterol	0	mg	Protein	4.12 g	Calcium	12.03 mg	29.04%	Calories from Total Fat
Total Fat	8.42	g	Sodium	46	mg	Vitamin A	0.0 RE	Iron	1.40 mg	2.08%	Calories from Saturated Fat
Saturated Fat	0.60	g	Carbohydrates	44.28	g	Vitamin A	0.3 IU	Water¹	17.02 g	*0.11%*	Calories from Trans Fat
Trans Fat¹	*0.03*	g	Dietary Fiber	3.98	g	Vitamin C	1.5 mg	Ash¹	0.23 g	67.86%	Calories from Carbohydrates
										6.32%	Calories from Protein

N/A - denotes a nutrient that is either missing or incomplete for an individual ingredient
* - denotes combined nutrient totals with either missing or incomplete nutrient data
¹ - denotes optional nutrient values

Table 1.3. Food barley standard reference panel for 2011-12 and 2012-13. See text for details on equipment and assays.
*Starch type determined from haplotype data.

Name	Source (2012)	Source (2013)	Row type	Hull type	Growth habit	Color	Starch type*	Protein 2012 (%)	Protein 2013 (%)	Grain β-glucan 2012 (%)	Kernel Hardness 2012 (SKCS)	Kernel Hardness 2013 (SKCS)
DZ100289	Hyslop Farm, Corvallis	Hyslop Farm, Corvallis	2-row	Hull-less	Winter	White	non-waxy	11.0	11.1	3.5	45.8	45.2
Karma	Hunton Farm, Junction City	Hyslop Farm, Corvallis	6-row	Hull-less	Spring	Purple	waxy	12.7	15.1	7.9	77.6	69.6
Streaker	Hyslop Farm, Corvallis	Hyslop Farm, Corvallis	6-row	Hull-less	Winter	Blue, brown, white	non-waxy	11.6	11.7	4.9	50.5	47.3
Tamalpais	Hunton Farm, Junction City	Hyslop Farm, Corvallis	6-row	Hull-less	Spring	White	non-waxy	12.4	14.8	8.8	77.4	78.2
Willamette Pearl	Hyslop Farm, Corvallis	Hyslop Farm, Corvallis	2-row	Hulled	Winter	White	waxy	12.1	10.8	6.5	61.2	44.2
Alba	Lewis Brown Farm, Corvallis	Hyslop Farm, Corvallis	6-row	Hulled	Winter	White	non-waxy	9.3	9.9	4.1	76.3	61.3
Full Pint	Lewis Brown Farm, Corvallis	Hyslop Farm, Corvallis	2-row	Hulled	Spring	White	non-waxy	9.2	11.5	4.6	72.0	35.9

Table 1.4. Mineral content for the standard reference panel for 2011-12. Values per 100 g. USDA standard values for hulled and pearled barley listed for comparison (no data available for Mn, Cu, or B).

Name	P mg	K mg	Ca mg	Mg mg	Mn mg	Cu mg	B mg	Zn mg
DZ100289	355	412	35	128	2.61	0.33	0.53	2.05
Karma	445	589	36	144	1.79	0.78	0.46	6.14
Streaker	467	557	70	133	3.47	0.80	0.67	2.80
Tamalpais	450	500	53	160	2.34	0.67	0.53	3.27
Willamette Pearl	502	579	69	156	2.20	1.00	0.61	3.68
Alba	306	514	38	130	1.51	0.67	0.76	2.05
Full Pint	404	751	40	157	1.93	1.33	0.99	4.50
USDA standard (hulled)	264	452	33	133				2.77
USDA standard (pearled)	221	280	29	79				2.13

Table 1.5. Mineral content for the standard reference panel for 2012-13. Values per 100 g. USDA standard values for hulled and pearled barley listed for comparison (no data available for Mn, Cu, or B).

Name	P mg	K mg	Ca mg	Mg mg	Mn mg	Cu mg	B mg	Zn mg	Fe mg
DZ100289	476	573	57	187	2.46	1.10	2.54	3.40	6.45
Karma	547	766	71	170	2.97	0.90	1.79	5.00	8.44
Streaker	412	500	71	148	2.56	0.84	1.41	3.60	6.77
Tamalpais	386	537	69	154	2.87	0.77	1.32	4.50	11.46
Willamette Pearl	371	590	75	143	2.87	0.71	1.13	3.30	6.92
Alba	306	552	54	139	2.46	0.65	1.04	2.60	5.17
Full Pint	403	599	65	168	3.48	0.97	1.04	4.20	11.70
USDA standard (hulled)	264	452	33	133				2.77	3.60
USDA standard (pearled)	221	280	29	79				2.13	2.50

Figure 4. Vitamin E content for the standard reference panel for 2011-12. aT = alpha-tocopherol; gT = gamma tocopherol; aT3 = alpha-tocotrienol; gT3 = gamma tocotrienol; dT3 = delta-tocotrienol. Alpha-tocopherol is the form required by humans, but all are potent lipid soluble antioxidants.

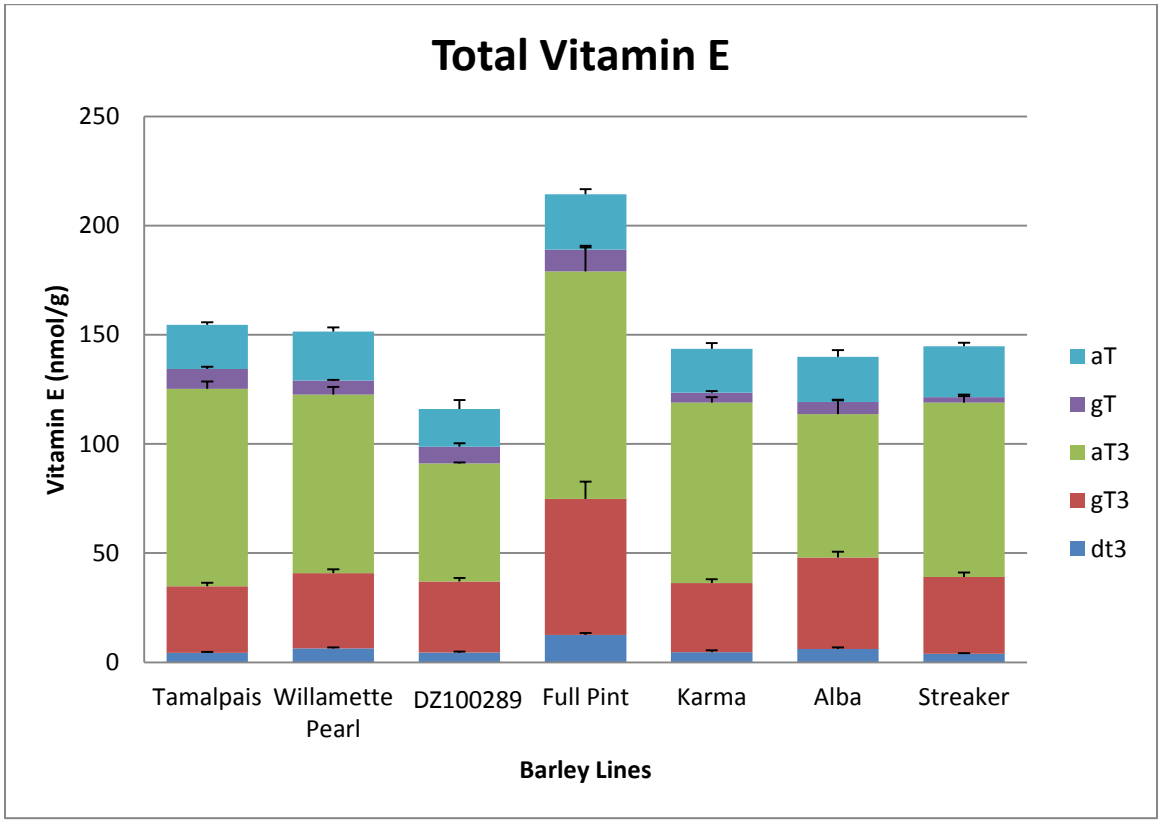
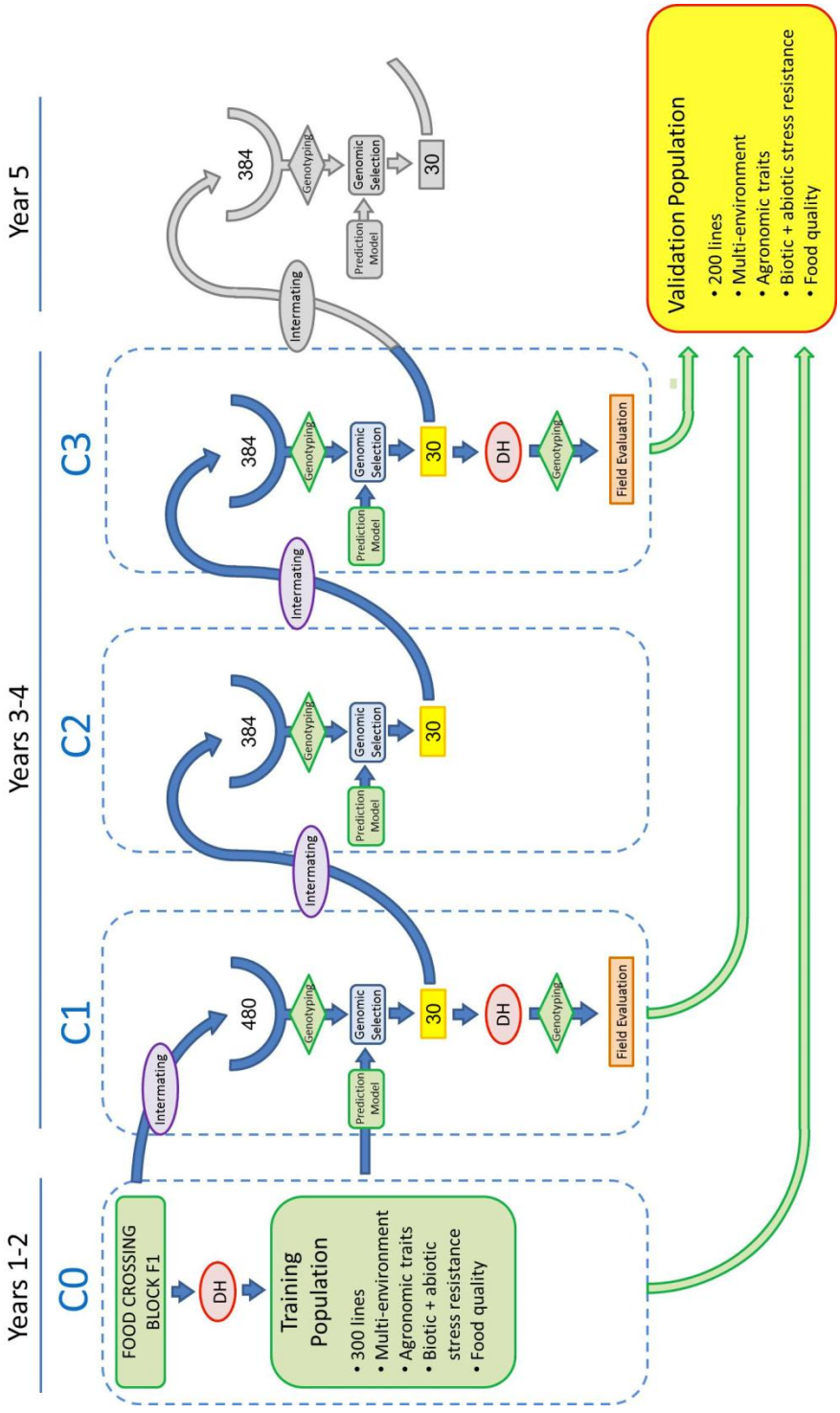


Figure 5. Doubled Haploid Genomic Selection breeding scheme. Germplasm described in Fig. 1 will be included in the training population.



Developing Winter Food Barley for the Pacific Northwest of the US

Brigid Meints, Alfonso Cuesta-Marcos, Andrew Ross, Chad Jackson, Juliet Marshall, Kevin Murphy, Scott Fisk, Teepakorn Kongraksawech, and Patrick Hayes.

Abstract:

Barley (*Hordeum vulgare* L.) is one of the oldest domesticated crops and has been cultivated for human consumption for thousands of years. However, most North Americans do not consume barley on a regular basis. In the last decade, there has been a renewed interest in barley production for human consumption. There are a number of quality traits that determine nutritional value and are useful for food processing. These include β-glucan, grain protein, kernel hardness, solvent retention capacity (SRC), and hull type. To determine the potential of winter growth habit food barley in the Pacific Northwest of the US, we developed and tested 14 advanced lines. The germplasm was developed via marker-assisted and phenotypic selection and included hulled lines with waxy starch and hull-less lines with normal starch. Grain yield, heading date, height, test weight, β-glucan, protein, kernel hardness, and SRC were measured on samples from three representative environments (dryland, irrigated, and high rainfall) over a two year period allowing for assessment of performance within and across locations, as well as genotype x environment interaction. Lines with waxy starch had significantly higher levels of β-glucan, harder kernels, and higher water retention capacity. Hull-less lines had, on average, lower yields than hulled lines but the difference was only 105 kg ha^{-1}. Hull-less selections from this germplasm array are in the variety release process and/or pipeline. Our

future food barley variety development will focus exclusively on hull-less types, due to the simplified processing and consumer interest in the nutritional benefits of whole grain.

Introduction:

Barley (*Hordeum vulgare* L.) is one of the oldest domesticated crops and has been cultivated for human consumption for thousands of years. However, most North Americans do not consume barley on a regular basis, and the majority of barley is grown for feed and malt. In 2006, the U.S. Food and Drug Administration approved a health claim for barley based on its potential for high levels of the soluble fiber, β-glucan, which has been shown to help reduce post-prandial glucose response, lower blood cholesterol levels, reduce insulin resistance, and reduce abdominal fat (AbuMweis et al., 2010; Bays et al., 2011; Behall et al., 2006; Casiraghi et al., 2006; Kim et al., 2009; Shimizu et al., 2008; Tiwari and Cummins, 2011). The health claim allows "foods containing barley to claim that they reduce the risk of coronary heart disease. Specifically, whole grain barley and dry milled barley products such as flakes, grits, flour, and pearled barley, which provide at least 0.75 grams of soluble fiber per serving" (21 CFR 101.81) (Ames and Rhymer 2008; National Barley Foods Council, 2003). Due to the health claim and reports that most North Americans do not get enough fiber in their diets (Slavin 2005), efforts have increased to breed new food barley varieties, characterize these varieties for food quality traits, and develop food

barley products (Baik and Ullrich 2008; Bhatty 1999; Newman and Newman 2008; Sullivan et al., 2012).

Grain β-glucan is influenced by both environmental and genetic factors. Anker-Nilssen et al. (2008) reported that barley grown in hotter and drier climates tends to have higher β-glucan than barley grown in wetter climates. Chutimanitsakun et al. (2013) also found that β-glucan content was significantly higher with increased daytime temperatures, even under irrigated conditions. There are qualitative and quantitative genetic components to grain β-glucan. In terms of qualitative variation, the recessive allele of the granule-bound starch synthase 1 (*GBSS1*) gene (also termed the "Waxy" (*WX*) locus) has a positive pleiotropic effect on grain β-glucan content (Szczodrak et al., 1992; Xue et al., 1997; Wood et al., 2003). Breeders have, therefore, selected for higher grain β-glucan by targeting the recessive (waxy, high amylopectin) allele (Patron et al., 2002; Islamovic et al., 2013). In terms of quantitative genetic variation for grain β-glucan, several QTL are reported in non-waxy, high amylose germplasm (Islamovic et al., 2013).

There are a number of other barley grain traits that are important, or potentially important for food uses. These include hull type, grain protein, kernel hardness, and solvent retention capacity (SRC). The hull-less trait (where the lemma and palea do not adhere to the hull) is recessive and determined by allelic variation at the *nud* locus. The *Nud* gene, controlled by an ERF family transcription factor, was cloned by Taketa et al. (2008). For malting barley, the adhering hull serves as a natural filtration device during the brewing process, but when breeding barley for

human consumption it is useful to breed for the hull-less trait, because the hull consists mainly of insoluble fiber (Baik et al., 2011) and the removal of the hull requires additional processing such as pearling. Pearling is the process of physically abrading the grain to remove the outer tissues including the hull, bran, and germ. Once a grain has been pearled, it no longer considered to be whole grain, which is defined as: "the intact, ground, cracked, or flaked caryopsis (kernel or seed), whose principal anatomical components—the starchy endosperm, germ, and bran—are present in the same relative proportions as they exist in the intact caryopsis" (Jones 2010). However, Choo et al. (2001) reported that the hull-less trait is associated with decreased yield, low seed weight, poor emergence, and short plant height compared with hulled types. After assessing food lines with spring growth habit under dryland conditions in the Pacific Northwest, Rey et al. (2009) also concluded that hulled lines had greater yield potential and increased vigor as compared to hull-less lines.

Grain protein is a quantitative trait controlled by QTL on all chromosomes, with the most important on chromosomes 2H, 4H, 5H, and 6H. Candidate genes were identified for two QTLs (*HvNAM1* on 6H and *HvNAM2* on 2H), which are homologs of genes controlling grain protein in wheat (Cai et al., 2013). Environment and growth practices can also have a significant impact on grain protein; increased availability of nitrogen, or heat stress due to drought, can increase protein levels (Zhang et al., 2001). Cai et al. (2013) stated that genotype is more important than environment in determining grain protein levels, whereas Zhang et al. (2001) argued the reverse. Grain protein is determined principally by the hordein storage proteins

found in the endosperm. The role of barley grain protein in human consumption has not been well studied or defined (Baik and Ullrich 2008).

Kernel hardness, defined as the "resistance of the kernel to deformation" (Turnbull and Rahmun 2002), is determined by endosperm texture and has a major effect on processing (milling and pearling) and the end-use of the grain. Harder kernels are more resistant to force and softer kernels are more easily damaged. Hordindolines and grain softness proteins have been mapped to the short arm of chromosome 5H in barley (Nair et al., 2010). This genome position is homeologous with the location of the same genes in wheat. However, unlike wheat, the biochemical basis of kernel hardness is not well understood in barley (Nair et al., 2010). In terms of processing, hard kernels result in a higher pearling yield, but require extra pearling time and greater energy. Hard kernels have a more densely packed endosperm, and during milling the starch particles remain trapped in the protein matrix, which results in larger particle sizes; soft kernels produce smaller particles (Nair et al., 2011). Similar to the market-classes that exist in wheat, barley end-use products may be determined by the softness or hardness of the initial grain input.

Solvent retention capacity (SRC) measures the capacity of a flour to absorb and retain four solvents: water, a lactic acid solution, a sodium carbonate solution, and a sucrose solution (reviewed by Kweon et al., 2011; AACC-International Approved Method 56-11.02). The basic principle of the test is that compatible solvents can swell polymeric networks. Different solvents emphasize swelling of different polymeric networks because of differences in solvent/polymer compatibility;

water swells all polymers in cereal flours (Kweon et al., 2011). SRC is commonly measured on wheat to assess end-use quality; however, there is little precedence for measuring SRC in barley (Slukova et al., 2012). In wheat, genotype plays a large role in the variation of values, although there is evidence of genotype x environment interactions (Guttieri and Souza, 2003).

There has been a renewed interest in producing barley for human consumption as a high fiber whole grain. In the Pacific Northwest of the US, where winter precipitation patterns prevail, winter and facultative growth habit barley varieties typically have a significant yield advantage over spring growth habit types. Therefore, we have focused our food barley breeding efforts on the former, using both marker assisted- and phenotypic selection (Chutimanitsakun et al., 2013). Advanced lines from were tested in the OFOOD (Oregon Food Barley) trial over a two-year period under dryland, irrigated, high rainfall, organic, and conventional conditions. A range of traits – agronomic, abiotic and biotic stress resistance, and grain quality – were measured on these advanced lines. Our objectives were to: (i) assess the agronomic performance of fall-sown barley food germplasm compared to check varieties, (ii) determine if there is a yield penalty associated with the hull-less trait, (iii) characterize food quality attributes, and (iv) assess the stability of agronomic and quality traits across different production environments and years.

Materials and methods:

Fourteen advanced generation food barley selections and two check varieties were included in the OFOOD trial. The checks were 'Alba': a hulled, non-waxy, winter, six-row, feed variety developed at Oregon State University (OSU) (Graebner et al., *submitted*) and 'Maja': a hulled, non-waxy, facultative, six-row, malt variety developed at OSU and released in 2006. The 14 food barley selections were developed using marker-assisted selection (MAS) and phenotypic selection (PS) as described by Chutimanitsakun et al. (2013). Briefly, the MAS project was designed to develop high β-glucan and winter growth habit germplasm via selection for specific alleles at the *WX* and *VRN-H2* loci. The parental germplasm for the MAS project included 'Luca' (two-row, normal starch, hulled, with winter growth habit, accessed from the Martonvasar Research Institute in Hungary), 'Merlin' and 'Waxbar' (two-row, waxy starch, hull-less, with spring growth habit, developed by Westbred Inc.), and 'Strider' (six-row, normal starch, hulled, with winter growth habit, released by the Oregon Agricultural Experiment Station in 1997). The PS germplasm was selected for the hull-less trait and agronomic performance in target environments. The parental germplasm consisted of normal starch types and included 'Strider' (see above), 'Doyce' (six-row, hull-less, with winter growth habit, developed at Virginia Polytechnic Institute) (Brooks *et al*., 2005), Maja (see above), and 'Legacy' (six-row, hulled, with spring growth habit, developed by Busch Agricultural Resources Inc.). Six of the advanced lines in the OFOOD trial were hull-less and non-waxy, seven were hulled and waxy, and one was hull-less and waxy (Table 2.1). Eight of the

entries (all waxy starch types) were derived by MAS; the remaining non-waxy starch types were derived by PS. All entries were selected for agronomic performance over multiple years and locations prior to inclusion in the OFOOD trial.

The OFOOD trial was grown over a two-year period (2011-2012 and 2012-2013) at eight locations using plot sizes, seeding rates, and management procedures appropriate for each location. In this report, we present data from the two years and three representative locations: Corvallis, OR (COR, representing high rainfall conditions); Pullman, WA (PUL, representing dryland conditions); and Aberdeen, ID (ABD, representing irrigated conditions). In COR 2012 and 2013, PUL 2012 and 2013, and ABD 2013 a randomized complete block (RCB) design with three replications was used. In ABD 2012 a RCB design with two replications was used. Grain yield, plant height, and heading date were measured on a plot basis. Grain yield and height measurements were replicated across all environments. Heading date was recorded across all locations for ABD 2012 and 2013, PUL 2012 and 2013, and COR 2013; only one replication was recorded in COR 2012. Test weight was measured on grain from all replications at ABD 2012 and 2013, and PUL 2012 and 2013; it was measured only on the first replication in COR 2012 and 2013. Food quality traits (grain β-glucan, protein, kernel hardness, and SRC) were measured on grain from a single replication from each location and in each year. Two technical replications were used when measuring kernel hardness and SRC. Winter survival was rated based on the visual assessment of the percentage of surviving plants on a plot basis at PUL and ABD on all replicates over the two years. No differential survival was

observed at COR. Resistance to barley stripe rust (incited by *Puccinia striiformis* f. sp. *hordei*) and scald (incited by *Rhynchosporium commune*) was rated by visual assessment of the percentage of leaf area affected by disease, on a plot basis, on all replicates at COR 2012 and on one replicate at COR 2013. These diseases were not observed at PUL and ABD.

For the measurement of grain β-glucan, whole grain samples were ground in a CleanMill 8000 (Newport Scientific, Sydney, Australia). The resulting flour was used to determine the mixed-linkage β-glucan percentage using the Megazyme enzymatic assay procedure (AACC Method 32-23.01; Megazyme International Ireland Ltd.) with the modified protocol developed by Hu and Burton (2008). Grain protein was measured using near infrared reflectance (NIR) spectroscopy (Infratec 1241 Grain Analyzer, Foss, Laurel, MD). Kernel hardness was measured using 300 kernels per sample on a SKCS 4100 (Perten Instruments, Springfield, IL) single kernel characterization system. Based on the report by Nair et al. (2010) that the hull has little effect on kernel hardness, we removed hulls from hulled selection in order to avoid clogging the SKCS machine. Grain samples from the hulled lines were pearled for 30 seconds using a Strong Scott Pearler (Seedboro Equipment Co., Chicago, IL). The kernel hardness of hull-less lines was measured using whole grain. Solvent Retention Capacity (SRC) was measured using the AACC-International Approved Method 56-11.02. Grain samples were milled on a laboratory hammer mill 3100 (Perten Instruments, Springfield, IL). Hull-less lines were milled from whole-grain and hulled lines were pearled for 30 seconds, as described for kernel hardness

assessment, and then milled. SRC is a composite method that uses four "solvents" to create a functionality fingerprint for a flour: water and three aqueous solutions, 50% w/w sucrose, 5% w/w sodium carbonate, and 5% w/w lactic acid. We found that with some barley samples and the sodium carbonate, lactic acid, and sucrose solutions, a complete pellet did not form after centrifugation. Therefore, we report only results for solvent retention capacity – water (referred to as SRC-W).

Combined analyses of variance were performed across locations and years for grain yield and height (replicated across all six environments) using the General Linear Models (GLM) procedure in SAS v9.3 (SAS Institute, Cary, NC, 2011). All effects were considered fixed in these analyses. For traits measured on only one or a subset of replications (heading date, test weight, β-glucan, protein, kernel hardness, and solvent retention capacity), years and/or locations were considered replicates. In order to assess genotype x location and genotype x year interactions, consistency plots and Additive Main effects and Multiplicative Interaction (AMMI) plots (Gauch 1988; Zobel et al., 1988) were created. Mean separation tests were based on F-protected LSD tests. Pearson's correlations were performed using the CORR procedure in SAS v9.3.

Results and Discussion:

Across six environments, significant differences among entries for all traits were observed (Table 2.2). Grain yield varied from 5701 to 8257 kg ha^{-1}; Alba (hulled, non-waxy) had the highest yield and OBADV10-14 (hull-less, non-waxy)

had the lowest yield. On average, hulled lines yielded 6458 kg ha^{-1}, whereas hull-less lines yielded 6353 kg ha^{-1}, a difference of only 105 kg ha^{-1}. Overall, the food barley germplasm was competitive with Maja, a six-row malting barley. Alba, which is a high-yielding feed variety, however, had a significant yield advantage over all experimental lines (Table 2.2). Alba is hulled however, and hull-less barley is more attractive for food purposes as it does not require pearling and meets the whole grain standard. On average, the barley hull is reported to account for 11-13% of total grain yield (Rey et al., 2009). When the yield of Alba is adjusted for hull (7266 kg ha^{-1}), the comparison with the average of the hull-less lines (6353 kg ha^{-1}) is more favorable, but food barley still has a ways to go to beat feed.

Our results are contrary to those of Choo et al. (2001) and Rey et al. (2009), who argued that the reduced vigor and lowered grain yield associated with the hull-less trait (even when the weight was adjusted to account for the hull) favors breeding and production of hulled food barley. Capo-chichi et al. (2012), reported that hull-less and hulled germplasm have similar seedling vigor. Although we did not measure seedling vigor in our experiments, our grain yield data are evidence that this sample of winter hull-less food barley germplasm has promise in terms of agronomic performance across a range of environments. Assessment of the impact of starch type on grain yield is confounded by the fact that only one hull-less experimental line (09OR-59) has waxy starch. This waxy starch line was among the lower yielding lines within the hull-less group, but the lowest yielding line had non-waxy starch.

Therefore, our data do not support a yield penalty for hull type or starch type in this sample of germplasm.

While Alba has a significantly higher yield than the other lines, it is critical to take care in assessing the significance of main effects and consider the genotype x environment (GxE) interactions. In the combined ANOVA of grain yield, all main effects (except for replication), two-way and three-way interactions were highly significant (Table 2.2). Location, year, and the location x year interaction accounted for the greatest portion of the variance. Genotype and the genotype interaction terms, while significant, were not as large. In the consistency plot (Figure 2.1), the basis of the significant genotype x location interaction is apparent; the median standard deviation of rank is quite high. This indicates that there were changes in rank across locations. Interestingly, two of the hull-less non-waxy accessions (09OR-86 and 09OR-89) were among the top-ranked entries for yield and had lower standard deviations that the highest yielding entry across locations – Alba (which is hulled). The AMMI plots of grain yield data are shown in Figures 2.2a and 2.2b. AMMI1 (Figure 2.2a), which plots yield performance by the first interaction principal component axis, shows that a number of lines that exhibit a positive interaction perform better under irrigated conditions (including Streaker and 09OR-86), while other lines perform better under dryland and high rainfall conditions. AMMI2 (Figure 2.2b) plots the interaction first and second principal components, which account for most of the GxE variance. The interaction first and second principal components account for 46% and 29%, respectively, of the total GxE variance for yield and both

are significant at $P<0.01$ according to the Gollob test (Gollob, 1967). According to the same test, other interaction principal components are not significant at $P<0.05$. Figure 2b describes more precisely which lines perform best under specific environmental conditions. This plot shows that the dryland (PUL) and high rainfall (COR) locations have a GxE effect in the same direction, though of different magnitude over the two years, but the irrigated location (ABD) tends to favor other lines, although depending on the year the direction of the interaction (positive or negative) is different. This indicates that even though there was a higher yield potential for certain lines under irrigated conditions, there was greater variability under irrigation between years than at the other locations.

Heights varied from 79.8 cm (09OR-55: hulled, waxy) to 98.3 cm (Alba: hulled, non-waxy) (Table 2.2). Hulled lines averaged 87.3 cm and hull-less lines averaged 90.9 cm. Choo et al. (2001) reported that the *nud* allele has a pleiotropic effect causing reduced plant height. However, our results show that hulled and hull-less lines were not significantly different in height. Alba was significantly taller than all but one experimental food line. Maja was not significantly different from eight of the experimental lines and significantly taller than six. Again, even with significant height differences between genotypes, it is important to consider GxE interactions. In the combined ANOVA of height, all main effects and two-way interactions were highly significant (Table 2.3). The three-way interaction was not significant. Location, year, and the location x year interaction accounted for a large portion of the variance. Genotype and the genotype interaction terms, while significant, were not as

important. Evidence of interactions can be seen in the consistency plot and AMMI plots (Figures 2.3, 2.4a, and 2.4b). In the consistency plot, the median standard deviation of rank is high, which indicates that genotypic ranks were very different based on location. Both checks fall below the median, along with 09OR-86, which indicates that these lines were consistently taller across all locations. In the AMMI plot for height, the interaction first and second principal components account for 54% and 23%, respectively, of the total GxE variance for height and both are the only significant components at $P<0.05$ according to the Gollob test (Gollob 1967). The AMMI plot reveals basis of the location x year interaction; in Figure 2.4b, the locations and years are spread across the quadrants, indicating that genotypes perform differently at the various locations depending on the year.

Mean values for heading date ranged between 119 and 142 Julian days (Table 2). 09OR-55 (hulled, waxy) flowered earliest, while 09OR-86 (hull-less, non-waxy) flowered latest. Based on the consistency plot (Figure 2.5), both checks and 09OR-86 consistently flowered the latest. There is an interesting trend observed in this plot: flowering time was more uniform in late lines, whereas early lines had much higher standard deviations of rank.

Mean test weight values ranged from 62.3 kg hL^{-1} (09OR-55: hulled, waxy) to 77.7 kg hL^{-1} (09OR-86: hull-less, non-waxy) (Table 2.2). Hulled lines averaged 65.3 kg hL^{-1}, significantly lower than hull-less lines, which averaged 75.5 kg hL^{-1}. This difference is expected and can be explained by the absence or presence of the hull, respectively. Figure 6 confirms this genotype x location interaction. All hull-less lines

are ranked higher than the hulled lines, but within the two classes, their ranks varied across locations.

Grain β-glucan content varied from 3.8% (Maja: hulled, non-waxy) to 6.4% (09OR-27: hulled, waxy) (Table 2.2). Entries with waxy starch had a significantly higher average β-glucan content (6.0%) compared to entries with normal starch, which averaged 4.4%. This is further evidence for the positive pleiotropic effect of the recessive allele at the *WX* locus on β-glucan. The difference we observed between the waxy and non-waxy classes corresponds with the values reported by Bhatty and Rossnagel (1998) and Fastnaught et al. (1996), where waxy barleys contained 6-8% β-glucan and non-waxy lines contained 4-6%. Based on the FDA health claim, in order to receive the daily recommended soluble fiber, a person needs to consume (per serving) at least 17g of steamed grain or 44g of bread made with 40% barley flour that contained 4.5% β-glucan. This amounts to a small side dish of steamed grain or two slices of bread per serving, which is manageable for most consumers. A consistency plot of mean ranks of β-glucan content by rank standard deviation showed that 09OR-59, the only hull-less waxy line had comparable ranks to the other waxy lines, but performed more consistently across locations than the other lines (Figure 2.7). Of the non-waxy lines, Streaker and the checks had low ranks, but were also consistent across locations. This is important for producers of whole grain products who need a consistent level of fiber.

Mean values for protein ranged from 11.3% to 13.8% (Table 2.2). Alba (hulled, non-waxy) had the lowest protein, while 09OR-28 (hulled, waxy) had the

highest. These values fall into the middle of the range of typical protein values found in barley (10-17%), but are equivalent to the values found in hull-less barley by Izydorczyk et al. (2000). A consistency plot showed that the checks and 09OR-86 had consistently low levels of protein across environments (Figure 2.8).

Kernel hardness values varied from 37.8 (OBADV10-14: hull-less, non-waxy) to 67.6 (Alba: hulled, non-waxy) SKCS HI units (Table 2). Waxy lines averaged 55.9 SKCS HI units, significantly higher than non-waxy lines, which averaged 47.4 SKCS HI units. Nair et al. (2010) reported a range of 30.1-91.9 SKCS HI units in 959 breeding lines. A subset of the 959 lines were examined for protein, amylose content, and β-glucan, but they found no significant correlations between kernel hardness and any of the other traits. However, Bhatty (1997) and Edney et al. (2002) did find evidence that endosperm texture is firmer as a result of waxy starch, which corresponds with our results. Kernel hardness ranks showed that Alba was the hardest grain and also consistently hard across locations (Figure 2.9). Streaker and 09OR-86 are both softer and have lower ranks, but Streaker was more consistent across locations. This is important because grain processors who are milling or flaking grain need to have a consistently soft or hard grain that will perform as expected.

Mean SRC-W values ranged from 98.2 (09OR-86: hull-less, non-waxy) to 146.9% (09OR-56: hulled, waxy) (Table 2.2). Lines with waxy starch had a significantly higher average (133.6%), than lines with normal starch, which averaged 102.2%. This genotype x location interaction is apparent in the consistency plot (Figure 2.10) where the waxy lines all fall on one side of the median and the non-

waxy lines fall on the other. There are differences in standard deviation of rank, which indicates that some lines, including Alba and 09OR-86, performed consistently across locations and others performed differently across environments. Solvent retention capacity is a test typically run on wheat, and there is only report for measuring it on barley (Slukova et al., 2012). We found that the lactic acid, sodium carbonate, and sucrose solutions did not give consistent results in our sample of germplasm. A compacted hydrated-flour pellet would not form after centrifugation, even with increased speed and time. There is no evidence in the literature that waxy starch in wheat causes problems with the different solvents. In the literature where SRC is performed on barley, there is also no mention of difficulties with protocol. Slukova et al. (2012) measured SRC on four barley samples (one was only barley bran) and obtained slightly lower percentages from the water test than we did (76-134%). The lines they measured had low protein content (between 6.7-10.9%) and low β-glucan (between 2.5-4.2%), which may be relevant because we found SRC-W to be positively correlated with both β-glucan and protein. More experimentation is required to adjust the protocol for the solvent retention capacity test, but once all four solvents can be used, this test will be very useful for classifying barley for specific end-use purposes. As new varieties are developed with different starch types and different degrees of kernel hardness, processors will need SRC data.

Mean barley stripe rust (BSR) percentages ranged from 0% (09OR-56, 09OR-62, and Maja) to 15% (09OR-28) (Table 2.4). All lines showed high levels of resistance, which is critical in high rainfall areas where BSR is prevalent and can

result in yield loss and lowered quality. The intensity of the stripe rust epidemics at Corvallis in 2011 and 2012 is apparent from the stripe rust severity of Thoroughbred, a susceptible line grown in an adjacent experiment, which was rated an average of 82.5% over the two years. Mean ratings for scald (on a 1-9 scale) ranged from 1.0 (Alba) to 6.5 (09OR-28) (Table 2.4). Most lines showed moderate susceptibility to the disease. Scald is not as devastating as BSR, but the necrotic lesions that form on foliar tissue can lead to yield loss and thin kernels (Garvin et al., 1997). Resistance to scald is difficult to achieve because the pathogen rapidly overcomes resistance genes (Brown et al., 1996; Garvin et al., 1997). Mean winter survival percentages ranged from 72.0 (09OR-59) to 97.4% (Alba) (Table 2.4). Differential winter survival was observed at PUL and ABD; all lines survived in COR both years. In our target environments in the Pacific Northwest a level of winterhardiness is essential and should be equivalent or greater than the checks. Figure 2.11 shows that the checks consistently have high levels of winterhardiness, as do some of the experimental lines. Other lines, including 09OR-86, are more inconsistent across locations. However, yield and winter survival were not correlated ($r = 0.16$, $P = 0.21$), which indicates that lines with intermediate levels of winterhardiness may tiller sufficiently to make up for plant mortality over the winter.

Across the six environments, significant positive correlations were observed between β-glucan and kernel hardness ($r = 0.40$, $P < .0001$), β-glucan and SRC-W ($r = 0.73$, $P < .0001$), kernel hardness and SRC-W ($r = 0.39$, $P < .0001$), and protein and SRC-W ($r = 0.50$, $P < .0001$) (Figure 2.12). These correlations confirm that this

germplasm fits the reported association of cereal β-glucans having high viscosity and water-binding capabilities, which allows for greater absorption of water (Izydorczyk and Dexter 2008; Lazaridou and Biliaderis 2007). Gamlath et al. (2008) reported significant positive correlations between kernel hardness and β-glucan content in malting barley (r = 0.873 and 0.764, $P<0.01$ over two years). Significant negative correlations were seen between test weight and β-glucan, kernel hardness, and SRC-W. The negative correlations with test weight were most likely confounded by the fact that all but one of the non-waxy lines were hull-less and all waxy lines were hulled.

Conclusions:

As interest in barley as a crop for human consumption grows, the demand for available agronomically sound varieties with good food qualities will increase. Our results from this experiment will help to meet needs of farmers, consumers, and processors. The overall grain yields achieved with this germplasm are much higher than those reported for spring barley germplasm by Rey et al. (2009). Additionally, despite some evidence that the hull-less trait is associated with lower yield and vigor, there were no significant differences between the hulled and hull-less classes in this germplasm. Therefore, our future food projects will be focused exclusively on breeding hull-less lines. This is due to an interest in the whole grain benefits and the processing difficulties that arise with pearling. We found that waxy starch played an important role in determining quality traits, including β-glucan, kernel hardness, and

SRC-W. Holtekjolen et al. (2008) reports that waxy starch can lead to difficulties in the baking process, if the appropriate amount of water is not used. Therefore, our efforts now focus on hull-less non-waxy types, with modest β-glucan levels. Streaker will be released in 2014, and 09OR-86 is a candidate for release.

Much progress has been made over the last decade in food barley breeding and characterization in different breeding programs around the world; our program will continue to focus on breeding nutritious and delicious barley varieties with good food quality that are adapted for the Pacific Northwest.

Acknowledgements:

The development of food barley and trialing of OFOOD is supported by the Oregon Agricultural Experiment Station, the Oregon Wheat Commission, the Idaho Barley Commission, Formula Grant no. 2013-31100-06041from the USDA National Institute of Food and Agriculture, and USDA-NIFA TCAP Project No. 2011-68002-30029. Thanks to all the collaborators who grew the OFOOD trial: Dr. Bradford Brown (University of Idaho; Parma, ID), Brook Brouwer and Dr. Steve Jones (Washington State University; Mount Vernon, WA), Mr. Mathias Kolding (Oregon State University emeritus; Hermiston, OR), and Dr. Steven Petrie and Karl Rhinhart (Oregon State University; Pendleton, OR). Thanks to Ann Corey, Juan Rey, Tanya Filichkin and Yada Chutimanitsakun for developing the germplasm that went into the OFOOD trial. Thanks to Laura Helgerson, Pierrette Castro, and Robyn Shepherd for their assistance measuring food quality traits.

References:

AACC International. Approved Methods of Analysis, 11th Ed. Method 56-11.02. Solvent Retention Capacity Profile. Approved June 3, 2009.AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-56-11.02

AACC International. Approved Methods of Analysis, 11th Ed. Method 32-23.01. β-glucan Content of Barley and Oats- Rapid Enzymatic Procedure. Approved November 3, 1999. AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-32-23.01

AbuMweis SS, Jew S, Ames NP (2010) β-glucan from barley and its lipid-lowering capacity: a meta-analysis of randomized, controlled trials. Eur J Clin Nutr 64: 1472-1480

Ames NP and Rhymer CR (2008) Issues surrounding health claims for barley. J Nutr 138: 1237S-1243S

Anker-Nilssen K, Sahlstrøm S, Knutsen SH, Holtekjølen AK, Uhlen AK (2008) Influence of growth temperature on content, viscosity and relative molecular weight of water-soluble β-glucans in barley (Hordeum vulgare L.) J Cereal Sci. 48(3): 670-677

Baik BK, Newman CW, Newman RK (2011) Food uses of barley. In: Ullrich SE (eds) Barley: Production, Improvement, and Uses. Wiley-Blackwell, 532-562

Baik BK and Ullrich SE (2008) Barley for food: characteristics, improvement, and renewed interest. J Cereal Sci. 48: 233-242

Bays H, Frestedt JL, Bell M, Williams C, Kolberg L, Schmelzer W, Anderson JW (2011) Reduced viscosity barley β-glucan versus placebo: a randomized controlled trial of the effects on insulin sensitivity for individuals at risk for diabetes mellitus. Nutr Metab. 8: 10

Behall KM, Scholfield DJ, Hallfrisch JG (2006) Barley β-glucan reduces plasma glucose and insulin responses compared with resistant starch in men. Nutr Res. 26: 644-650

Bhatty RS (1997) Milling of Regular and Waxy Starch Hull-less Barleys for the Production of Bran and Flour 1. Cereal Chem. 74(6): 693-699

Bhatty RS (1999) The potential of hull-less barley. Cereal Chem. 76(5): 589-599

Bhatty RS and Rossnagel BG (1998) Comparison of pearled and unpearled Canadian and Japanese barleys. Cereal Chem. 75(1): 15-21

Brooks WS, Griffey CA, Price AM, Pridgen TH, Rohrer WL (2005) Registration of 'Doyce' Hulless Barley the ventral furrow. Crop Sci. 45: 791-792

Brown AHD, Burdon JJ, Garvin DF, Abbott DC, Read BJ (1996) The effect of combining scald resistance genes on disease levels, yield and quality traits in barley. Theor Appl Genet. 93(3): 361-366

Cai S, Yu G, Chen X, Huang Y, Jiang X, Zhang G, Jin X (2013) Grain protein content variation and its association analysis in barley. BMC Plant Biol. 13(1): 35

Capo-chichi L, Kenward K, Nyachito J, Anyia A (2012) Nud locus and the effects on seedling vigour related traits for genetic improvement of hulless barley. Journal of Plant Science and Molecular Breeding. 1(1): 2

Casiraghi MC, Garsetti M, Testolin G, Brighenti F (2006) Post-prandial responses to cereal products enriched with barley β-glucan. J Am Coll Nutr. 25: 313-320

Choo TM, Ho KM, Martin RA (2001) Genetic analysis of a hulless × covered cross of barley using doubled-haploid lines. Crop Sci. 41(4): 1021-1026

Chutimanitsakun Y, Cuesta-Marcos A, Chao S, Corey A, Filichkin T, Fisk S, Kolding M, Meints B, Ong YL, Rey JI, Ross AS, Hayes PM (2013) Application of marker-assisted selection and genome-wide association scanning to the development of winter food barley germplasm resources. Plant Breeding. 132(6): 563-570

Edney MJ, Rossnagel BG, Endo Y, Ozawa S, Brophy M (2002) Pearling Quality of Canadian Barley Varieties and their Potential Use as Rice Extenders. J Cereal Sci. 36(3): 295-305

Fastnaught CE, Berglund PT, Holm ET, Fox GJ (1996) Genetic and environmental variation in β-glucan content and quality parameters of barley for food. Crop Sci. 36(4): 941-946

Gamlath J, Aldred GP, Panozzo JF (2008) Barley (1→ 3; 1→ 4)-β-glucan and arabinoxylan content are related to kernel hardness and water uptake. J Cereal Sci. 47(2): 365-371

Garvin DF, Brown AHD, Burdon JJ (1997) Inheritance and chromosome locations of scald-resistance genes derived from Iranian and Turkish wild barleys. Theor Appl Genet. 94(8): 1086-1091

Gauch HG (1988) Model selection and validation for yield trials with interaction. Biometrics. 44: 705–715

Gollob HF (1968) A statistical model which combines features of factor analytic and analysis of variance techniques. Psychometrika. 33: 73–115

Guttieri MJ and Souza E (2003) Sources of variation in the solvent retention capacity test of wheat flour. Crop Sci. 43(5): 1628-1633

Holtekjølen AK, Olsen HHR, Færgestad EM, Uhlen AK, and Knutsen SH (2008) Variations in water absorption capacity and baking performance of barley varieties with different polysaccharide content and composition. Food Sci Technol-LEB (10): 2085-2091

Hu G and Burton C (2008) Modification of standard enzymatic protocol to a cost-efficient format for mixed-linkage (1→3,1→4)-β-d-glucan measurement. Cereal Chem. 85: 648-653

Islamovic E, Obert DE, Oliver RE, Harrison SA, Ibrahim A, Marshall JM, Miclaus KJ, Hu G, Jackson EW (2013) Genetic dissection of grain β-glucan and amylose content in barley (*Hordeum vulgare* L.). Mol Breeding. 31: 15-25

Izydorczyk MS, Storsley J, Labossiere D, MacGregor AW, Rossnagel BG (2000) Variation in total and soluble β-glucan content in hulless barley: effects of thermal, physical, and enzymic treatments. J Agr Food Chem. 48(4): 982-989

Izydorczyk MS and Dexter JE (2008) Barley β-glucans and arabinoxylans: molecular structure, physicochemical properties, and uses in food products–a review. Food Res Int. 41(9): 850-868

Jones JM (2010) Dietary Fibre‘s Co-Passengers: Is It the Fibre or the Co-Passengers pp 365-378. In:. Van Der Kamp J.W., Jones J, Mccleary B, Topping DJ (eds.) Dietary Fibre: New Frontiers for Food and Health. Wageningen Academic Publishers: Wageningen, The Netherlands

Kim H, Stote KS, Behall KM, Spears K, Vinyard B, Conway JM (2009) Glucose and insulin responses to whole grain breakfasts varying in soluble fiber, β-glucan. Eur J Nutr. 48: 170-175

Kweon M, Slade L, Levine H (2011) Solvent Retention Capacity (SRC) Testing of Wheat Flour: Principles and value in predicting flour functionality in different wheat-based food processes and in wheat breeding-A review. Cereal Chem. 88(6): 537-552

Lazaridou A, and Biliaderis CG (2007). Molecular aspects of cereal β-glucan functionality: Physical properties, technological applications and physiological effects. J Cereal Sci. 46(2): 101-118

Nair S, Ullrich SE, Blake TK, Cooper B, Griffey CA, Hayes PM, Hole DJ, Horsley RD, Obert DE, Smith KP, Muehlbauer GJ, Baik, B-K (2010) Variation in kernel hardness and associated traits in US barley breeding lines. Cereal Chem. 87: 461-466

Nair S, Ullrich SE, Baik BK (2011) Association of barley kernel hardness with physical grain traits and food processing parameters. Cereal Chem. 88(2): 147-152

National Barley Foods Council (2003) Petition for unqualified health claim: Barley β-glucan soluble fiber and barley products containing β-glucan soluble fiber and coronary heart disease. Submitted to the FDA on September 25, 2003.

Newman RK and Newman CW (2008) Barley for food and health: Science, technology, and products. John Wiley & Sons.

Patron NJ, Smith AM, Fahy BF, Hylton CM, Naldrett MJ, Rossnagel BG, Denyer K (2002) The altered pattern of amylose accumulation in the endosperm of low-amylose barley cultivars is attributable to a single mutant allele of granule-bound starch synthase I with a deletion in the 5′-non-coding region. Plant Physiol. 130(1): 190-198

Rey JI, Hayes PM, Petrie SE, Corey A, Flowers M, Ohm JB, Ong C, Rhinhart K, Ross AS (2009) Production of dryland barley for human food: Quality and agronomic performance. Crop Sci. 49(1): 347-355

Shimizu C, Kihara M, Aoe S, Araki S, Ito K, Hayashi K, Watari J, Sakata Y, Ikegami S (2008) Effect of high β-glucan barley on serum cholesterol concentrations and visceral fat area in Japanese men - A randomized, double-blinded, placebo-controlled trial. Plant Food Hum Nutr. 63: 21-25

Slavin JL (2005) Dietary fiber and body weight. Nutrition. 21: 411-418

Slukova M, Velebna N, Krejcirova L, Honcu I, Budilova E (2012) The Advanced Approaches to Nutritional and Breadmaking Quality of Wheat, Barley and Rye Flour. Journal of Food Science and Engineering. 2(4): 218-226

Sullivan P, Arendt E, Gallagher E (2013) The increasing use of barley and barley by-products in the production of healthier baked goods. Trends Food Sci Tech. 29(2): 124-134 (http://www.sciencedirect.com/science/article/pii/S0924224412002130)

Szczodrak J, Czuchajowska Z, Pomeranz Y (1992) Characterization and estimation of barley polysaccharides by near-infrared spectroscopy. II: Estimation of total β-D-glucans. Cereal Chem. 69(4): 419-423

Taketa S, Amano S, Tsujino Y, Sato T, Saisho D, Kakeda K, Takeda K (2008) Barley grain with adhering hulls is controlled by an ERF family transcription factor gene regulating a lipid biosynthesis pathway. P Natl Acad Sci USA. 105(10): 4062-4067

Tiwari U and Cummins E (2011) Meta-analysis of the effect of β-glucan intake on blood cholesterol and glucose levels. Nutrition. 27: 1008-1016

Turnbull KM and Rahman S (2002) Endosperm texture in wheat. J Cereal Sci. 36(3): 327-337

Wood PJ, Weisz J, Beer MU, Newman CW, Newman RK (2003) Structure of (1→ 3)(1→ 4)-β-d-glucan in waxy and nonwaxy barley. Cereal Chem. 80(3): 329-332

Xue Q, Wang L, Newman RK, Newman CW, Graham H (1997) Influence of the hulless, waxy starch and short-awn genes on the composition of barleys. J Cereal Sci. 26(2): 251-257

Zhang G, Chen J, Wang J, Ding S (2001) Cultivar and environmental effects on (1→ 3, 1→ 4)-β-D-glucan and protein content in malting barley. J Cereal Sci. 34(3): 295-301

Zobel RW, Wright MJ, Gauch HG (1988) Statistical analysis of a yield trial. Agron J. 80: 388–393

Table 2.1. Pedigree, row type, hull type, and starch type for all lines in the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.

Line	Pedigree	Row type	Hull type	Starch type
Streaker	Maja/Legacy, F1//Maja///Doyce	Six-row	Hull-less	Normal
OBADV10-13	Strider/Doyce	Six-row	Hull-less	Normal
OBADV10-14	Strider/Doyce	Six-row	Hull-less	Normal
09OR-59	Strider/Merlin, F1//Strider	Six-row	Hull-less	Waxy
09OR-70	Maja/Legacy, F1//Maja///Doyce	Six-row	Hull-less	Normal
09OR-86	Strider/Doyce	Six-row	Hull-less	Normal
09OR-89	Strider/Doyce	Six-row	Hull-less	Normal
09OR-27	Luca/Merlin, F1//Luca	Two-row	Hulled	Waxy
09OR-28	Luca/Merlin, F1//Luca	Two-row	Hulled	Waxy
09OR-31	Luca/Merlin, F1//Luca	Two-row	Hulled	Waxy
09OR-51	Luca/Waxbar, F1//Luca	Two-row	Hulled	Waxy
09OR-55	Strider/Merlin, F1//Strider	Six-row	Hulled	Waxy
09OR-56	Strider/Merlin, F1//Strider	Six-row	Hulled	Waxy
09OR-62	Strider/Merlin, F1//Strider	Six-row	Hulled	Waxy
Alba	Strider/Orca	Six-row	Hulled	Normal
Maja	Strider/88Ab536	Six-row	Hulled	Normal

Table 2.2. Means of grain yield, heading date, height, test weight, β-glucan, protein, kernel hardness, and solvent retention capacity of water (SRC-W) of the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.
*Based on field experiment replication. All other traits use environments as replications.

Line	Yield	Height	Heading Date	Test Weight	β-glucan	Protein	Kernel hardness	SRC-W
	kg ha^{-1}	cm	Julian days	kg hL^{-1}	% (w/w)	%	SKCS HI units	%
Streaker	6512	91.3	134	74.8	4.2	12.7	44.0	102.5
OBADV10-13	6114	92.3	135	77.4	4.6	11.6	39.3	101.5
OBADV10-14	5701	91.2	137	75.8	4.2	11.7	37.8	98.6
09OR-59	5837	84.9	133	73.1	5.9	13.7	50.7	129.8
09OR-70	6498	87.4	132	72.5	4.8	13.2	57.2	110.4
09OR-86	6758	95.3	142	77.7	4.1	11.4	42.9	98.2
09OR-89	7049	93.7	141	77.0	4.2	11.7	43.8	101.9
09OR-27	6274	93.1	134	66.6	6.4	12.1	58.1	142.9
09OR-28	5804	83.8	131	66.2	6.3	13.8	53.2	130.7
09OR-31	6180	88.1	130	66.2	6.3	13.3	51.6	137.7
09OR-51	6705	84.0	132	67.6	6.0	13.1	47.4	131.9
09OR-55	6411	79.8	119	62.3	5.7	12.1	61.7	123.9
09OR-56	7053	84.3	133	63.7	6.1	12.7	66.0	146.9
09OR-62	6777	83.0	132	64.6	5.5	12.6	58.6	125.0
Alba	8257	98.3	141	66.0	4.4	11.3	67.6	108.4
Maja	6856	91.1	136	63.9	3.8	11.7	46.8	102.1
Mean	6549	88.8	134	69.7	5.2	12.4	51.7	118.3
LSD (P = 0.05)	535*	4.6*	7	2.3	0.6	1.1	5.9	11.4
CV%	12.2	7.6	4.7	2.9	10.3	7.8	9.8	8.4

Table 2.3. Estimates of variance components for grain yield and height for the OFOOD trial at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.
*Significant at $P < 0.05$.
**Significant at $P < 0.01$.
‡ns, not significant.

Component of variance	df	Grain yield	Height
Genotype	15	7651024**	449.9**
Rep (Location x Year)	11	598746ns‡	94.8*
Location	2	230291247**	6805.0**
Year	1	16787710**	2345.2**
Location x Year	2	44361578**	4874.2**
Genotype x Location	30	2806094**	189.4**
Genotype x Year	15	285128*	162.4**
Genotype x Location x Year	30	1812859**	65.4ns
Error	165	622844	45.8

Figure 2.1. Consistency plot of mean rank by standard deviation of rank for grain yield at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

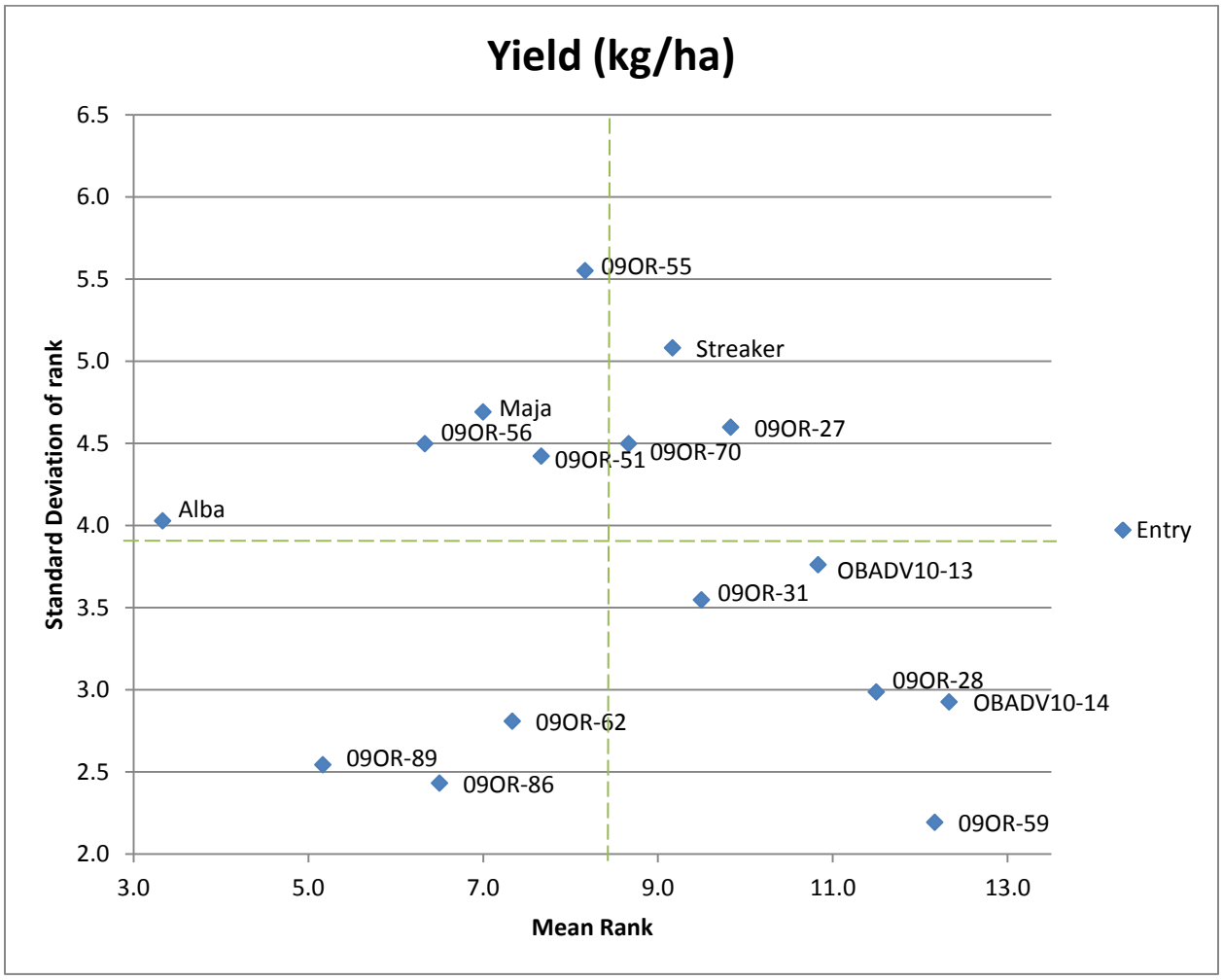


Figure 2.2a. AMMI1 plot for grain yield at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013.

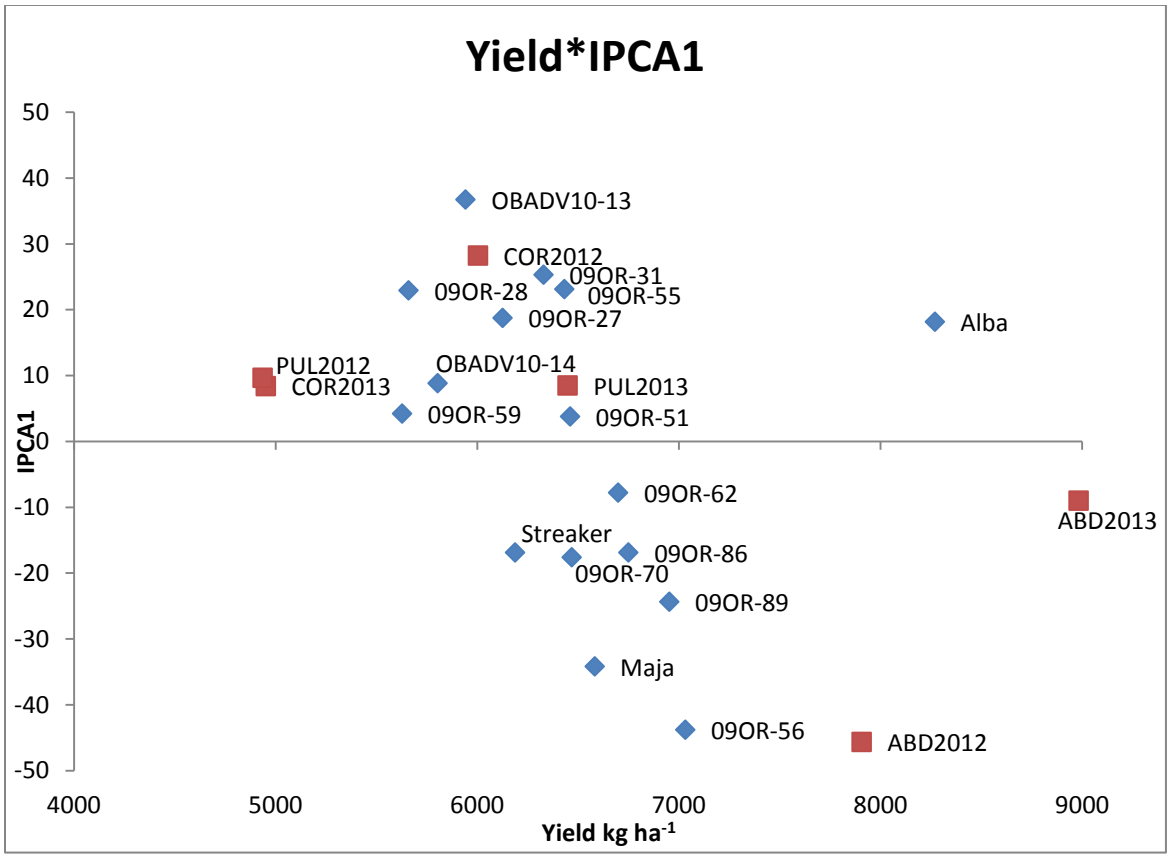


Figure 2.2b. AMMI2 plot for grain yield at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013.

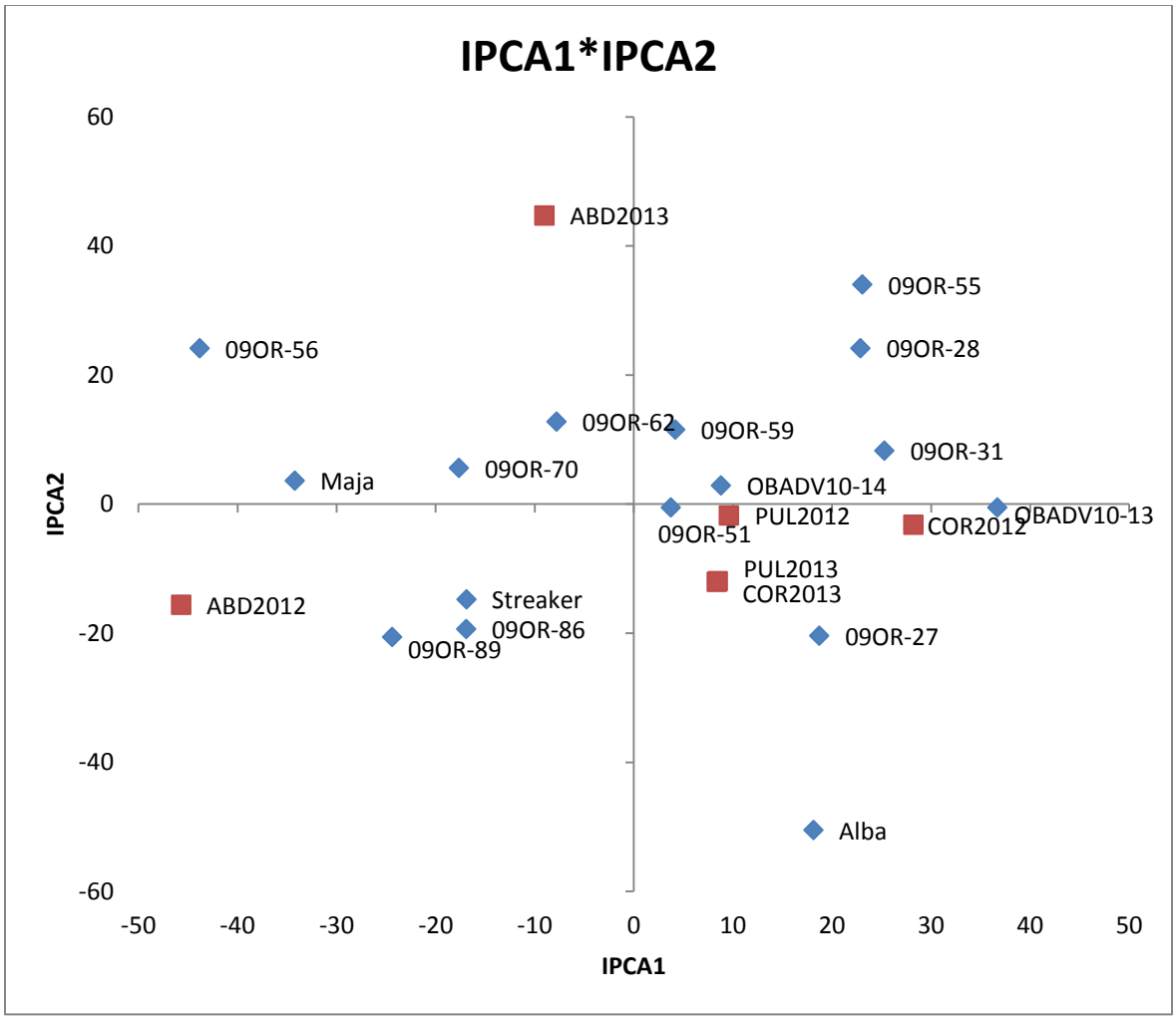


Figure 2.3. Consistency plot of mean rank by standard deviation of rank for height at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

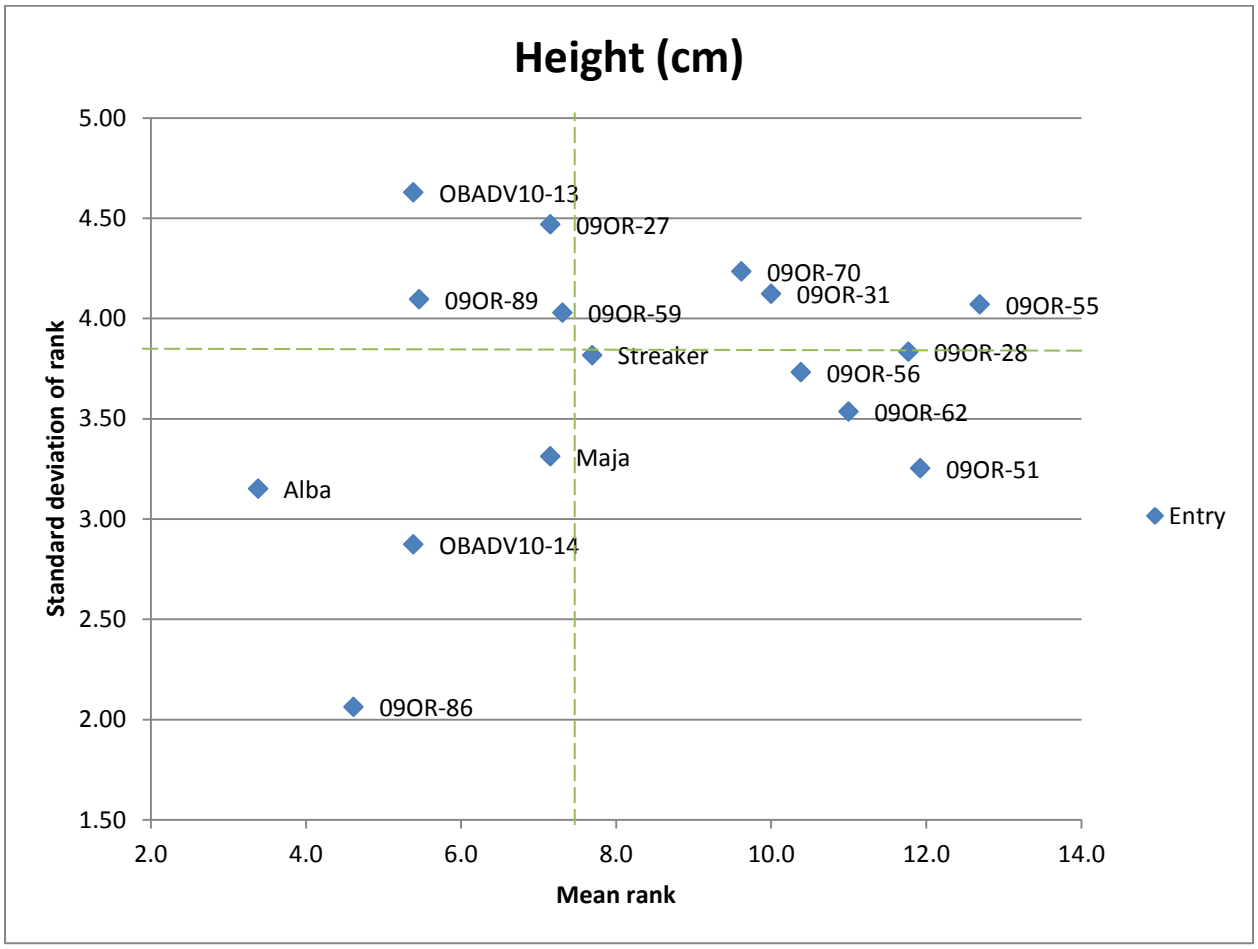


FIgure 2.4a. AMMI1 plot for plant height at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013.

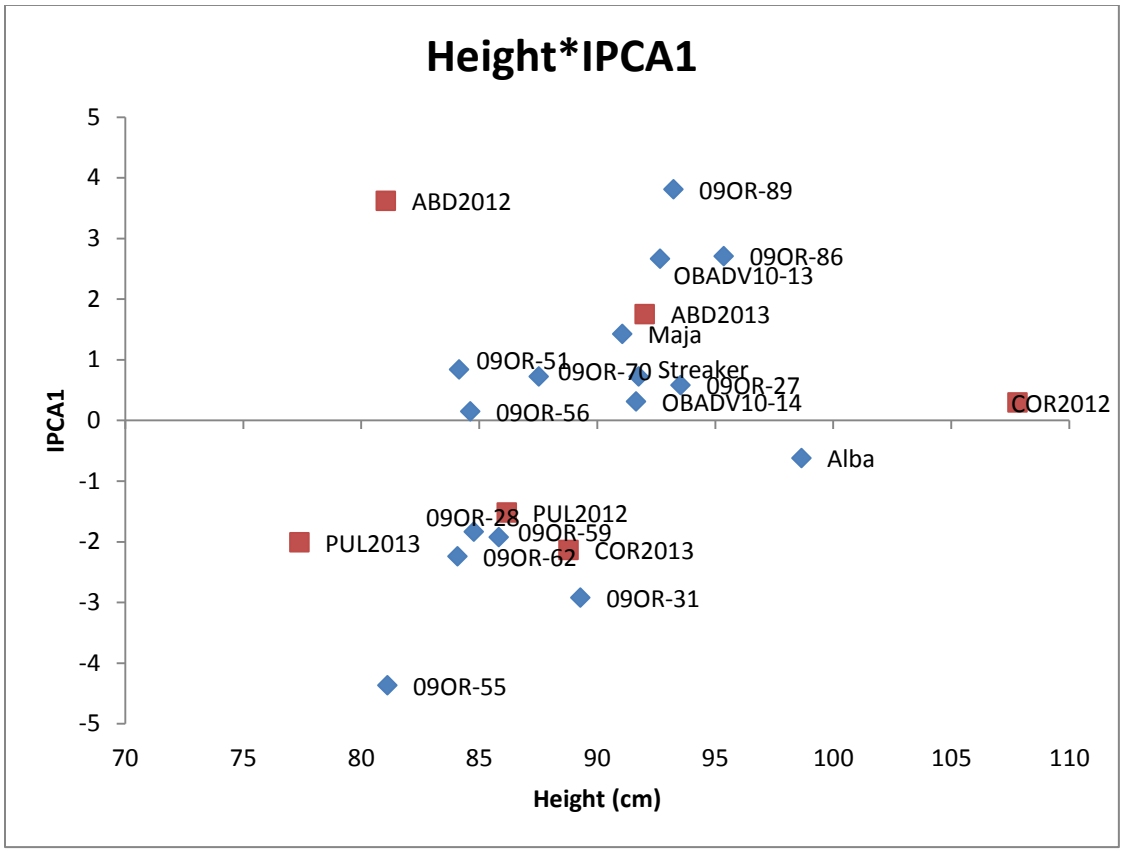


Figure 2.4b. AMMI2 plot for plant height at Corvallis, OR (COR); Pullman, WA (PUL); and Aberdeen, ID (ABD) in 2012 and 2013.

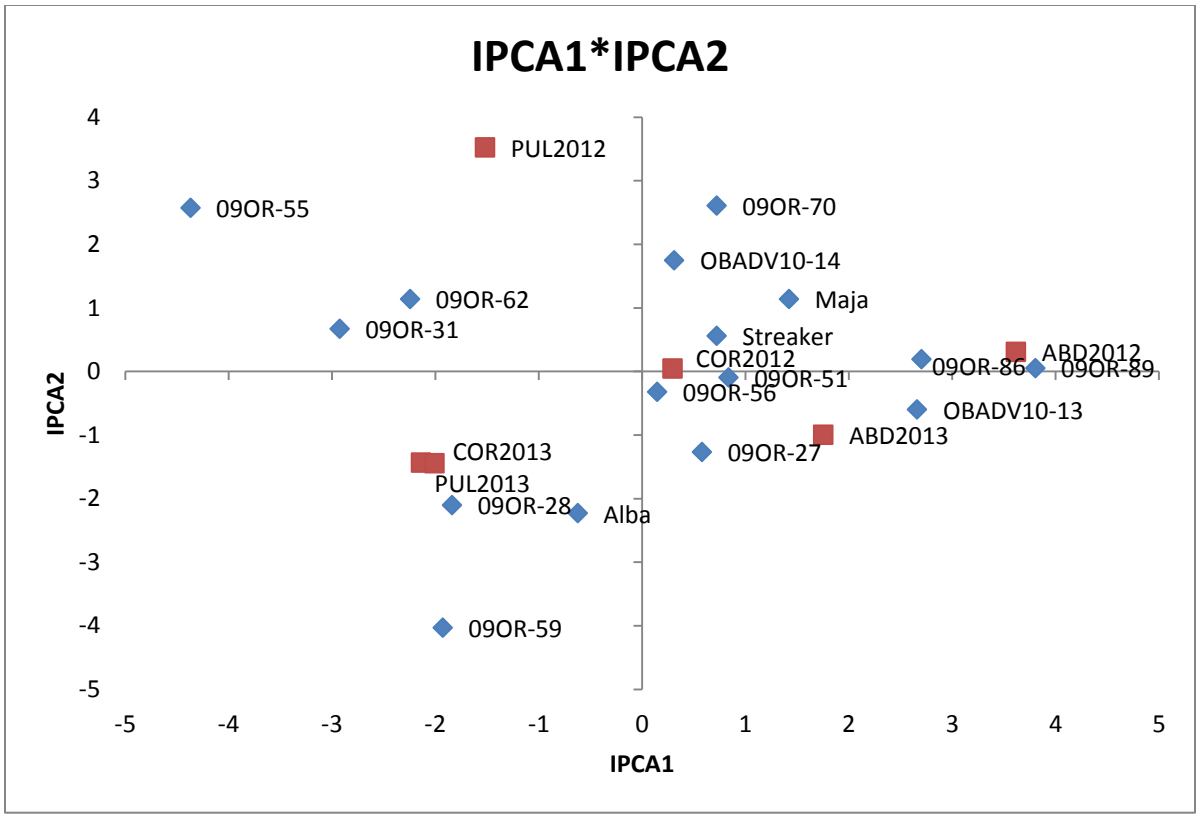


Figure 2.5. Consistency plot of mean rank by standard deviation of rank for heading date at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

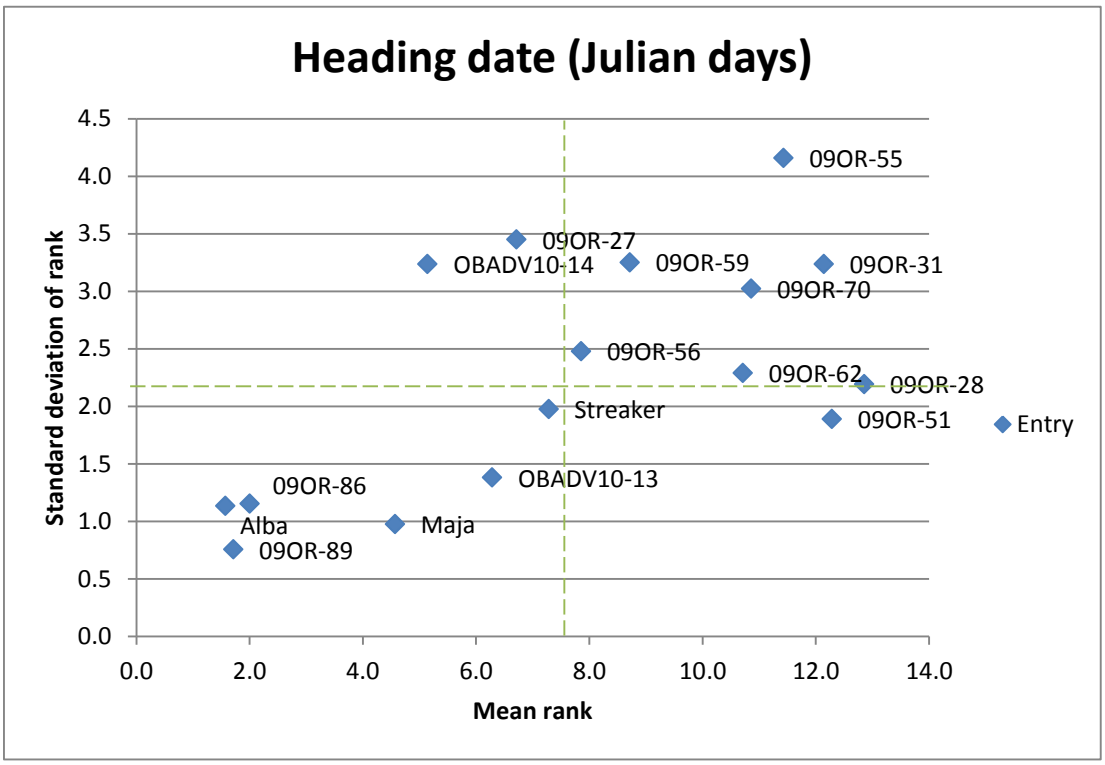


Figure 2.6. Consistency plot of mean rank by standard deviation of rank for test weight at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

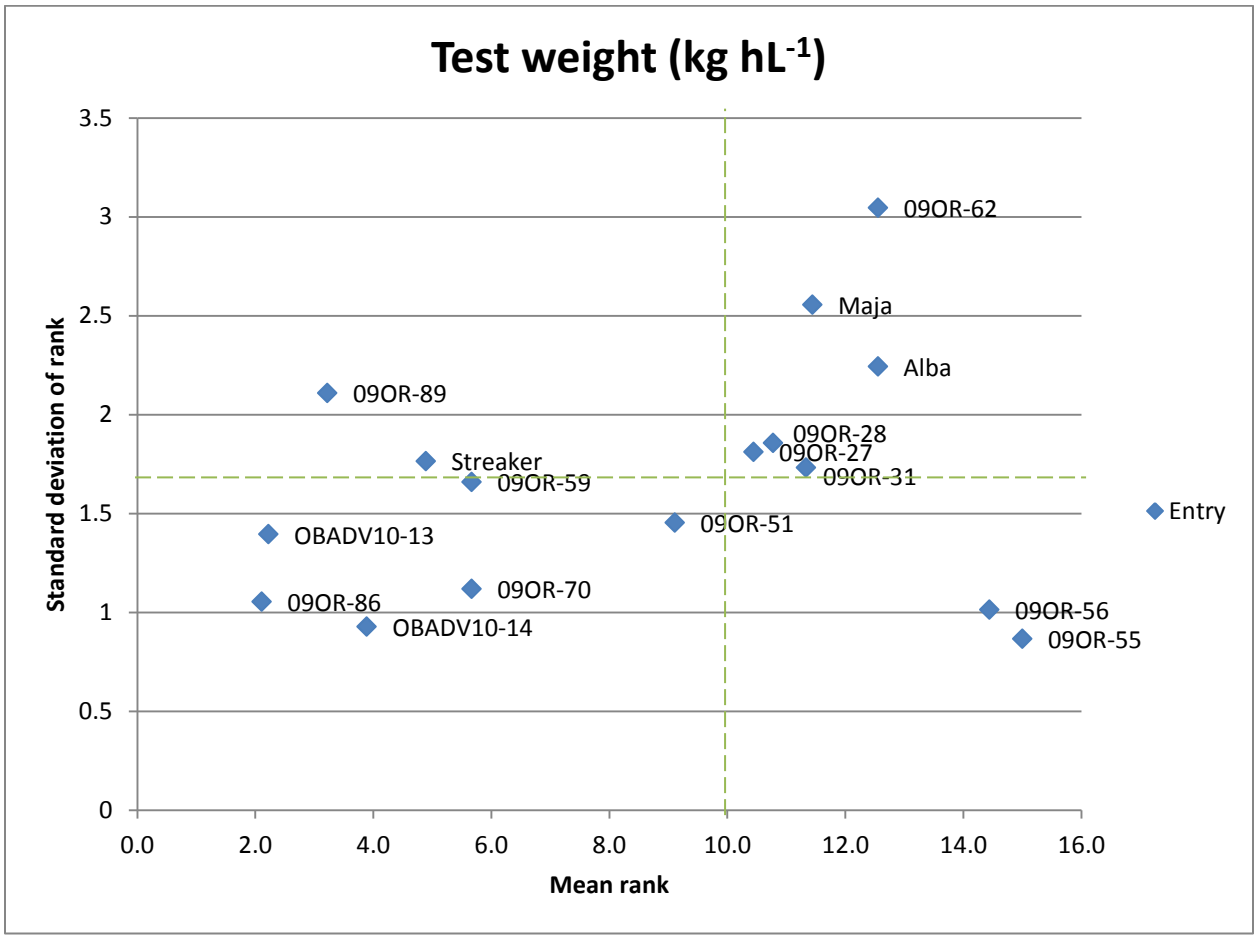


Figure 2.7. Consistency plot of mean rank by standard deviation of rank for β-glucan at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

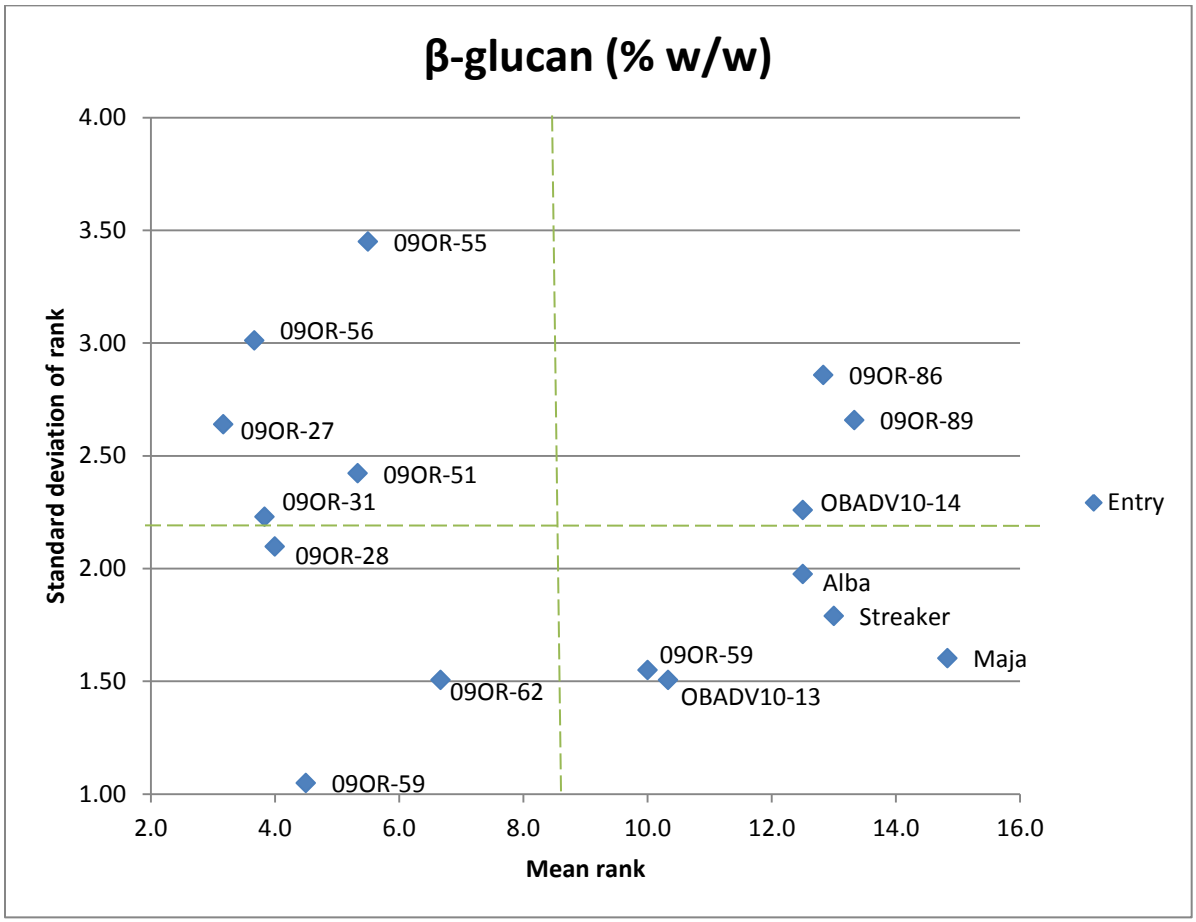


Figure 2.8. Consistency plot of mean rank by standard deviation of rank for protein at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

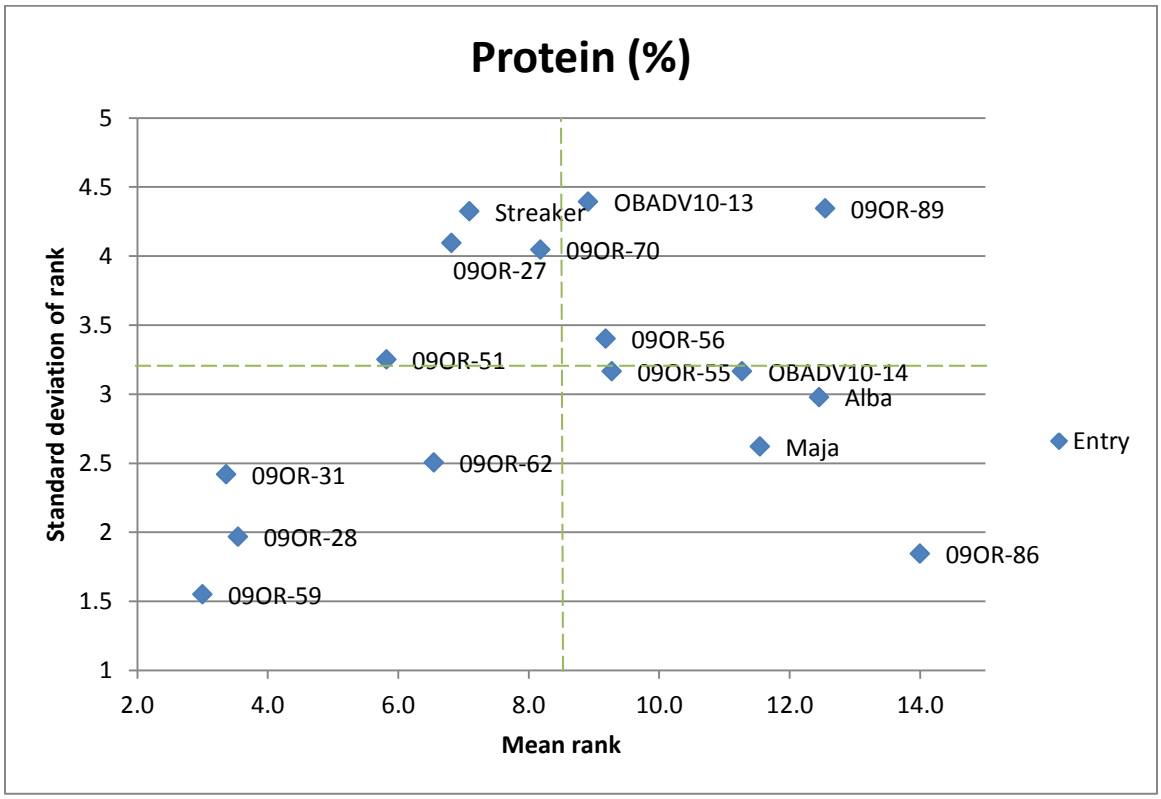


Figure 2.9. Consistency plot of mean rank by standard deviation of rank for kernel hardness at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

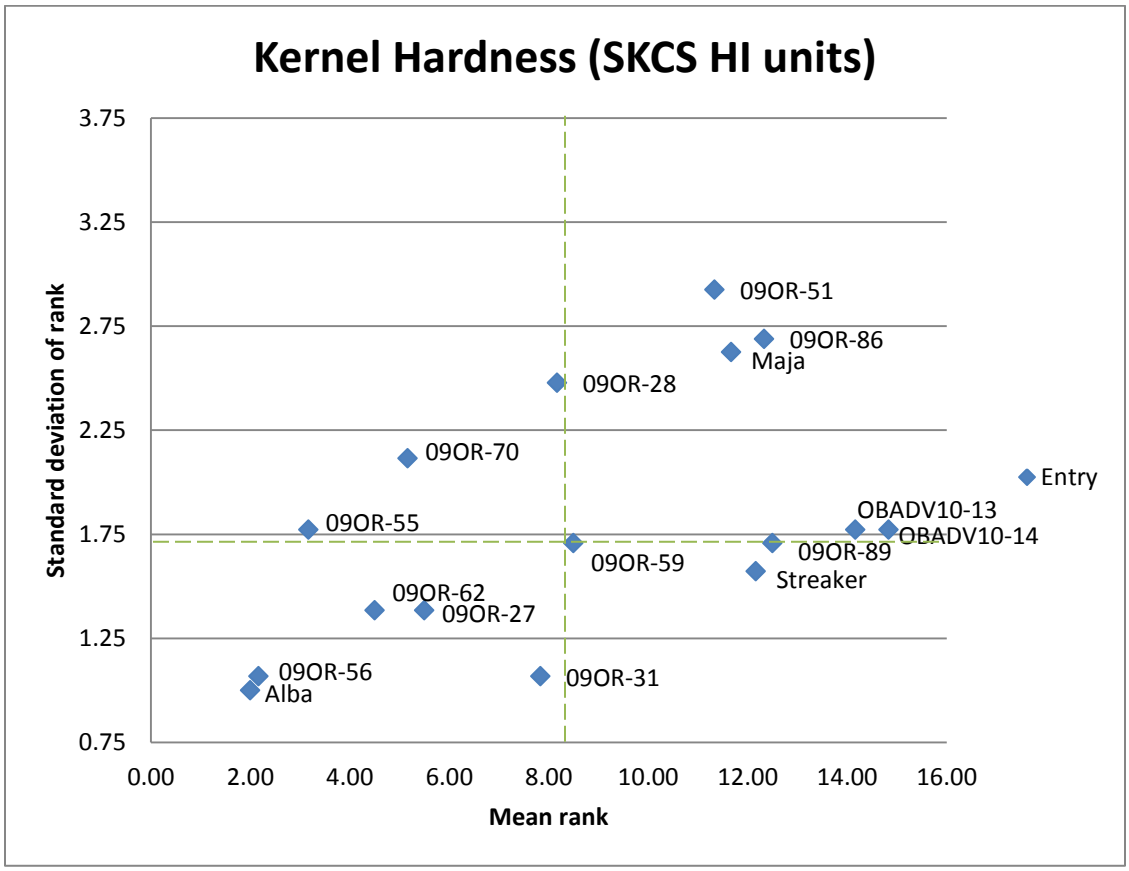


Figure 2.10. Consistency plot of mean rank by standard deviation of rank for solvent retention capacity of water (SRC-W) at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

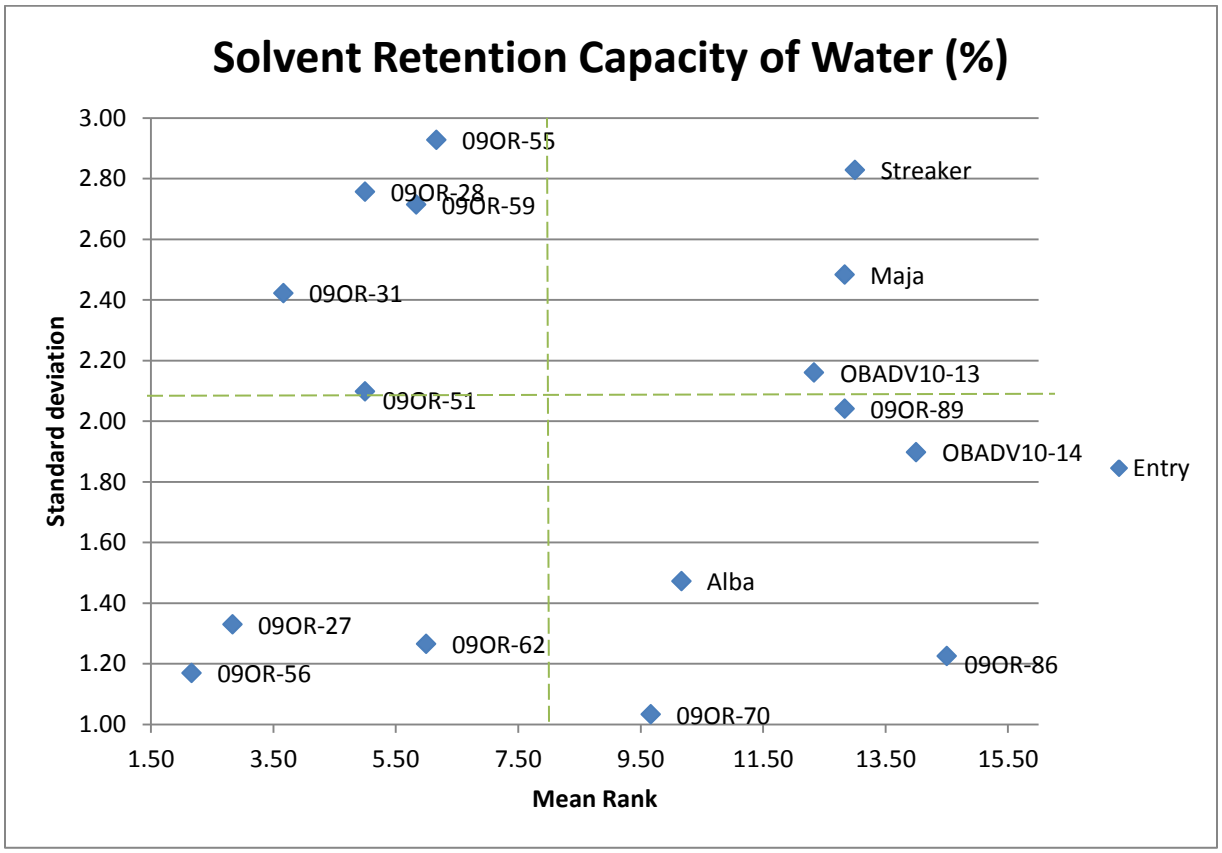


Table 2.4. Means for barley stripe rust (BSR) and scald for the OFOOD trial at Corvallis, OR in 2012 and 2013. Means for winter survival for the OFOOD trial at Pullman, WA and Aberdeen, ID in 2012 and 2013.
*1-9 scale where 1=most resistant and 9=most susceptible
‡Based on field experiment replication. Disease notes based on replication by year.

Line	BSR	Scald	Winter survival
	%	1-9 scale*	%
Streaker	10.0	6.3	90.5
OBADV10-13	10.5	4.0	79.3
OBADV10-14	10.0	3.5	87.5
09OR-59	5.0	4.5	72.0
09OR-70	10.0	3.8	92.3
09OR-86	5.0	3.0	76.2
09OR-89	5.0	3.0	81.0
09OR-27	10.0	5.5	90.0
09OR-28	15.0	6.5	94.7
09OR-31	12.5	6.0	95.6
09OR-51	12.5	4.5	91.9
09OR-55	10.0	3.5	80.5
09OR-56	0.0	4.5	87.3
09OR-62	0.0	4.0	93.8
Alba	5.0	1.0	97.4
Maja	0.0	4.5	91.4
Mean	7.5	4.3	87.6
Number of env.	2	2	4
LSD	13.0	2.0	8.1‡
CV	80.7	22.0	10.9

Figure 2.11. Consistency plot of mean rank by standard deviation of rank for winter survival at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013. Dashed lines indicate median values.

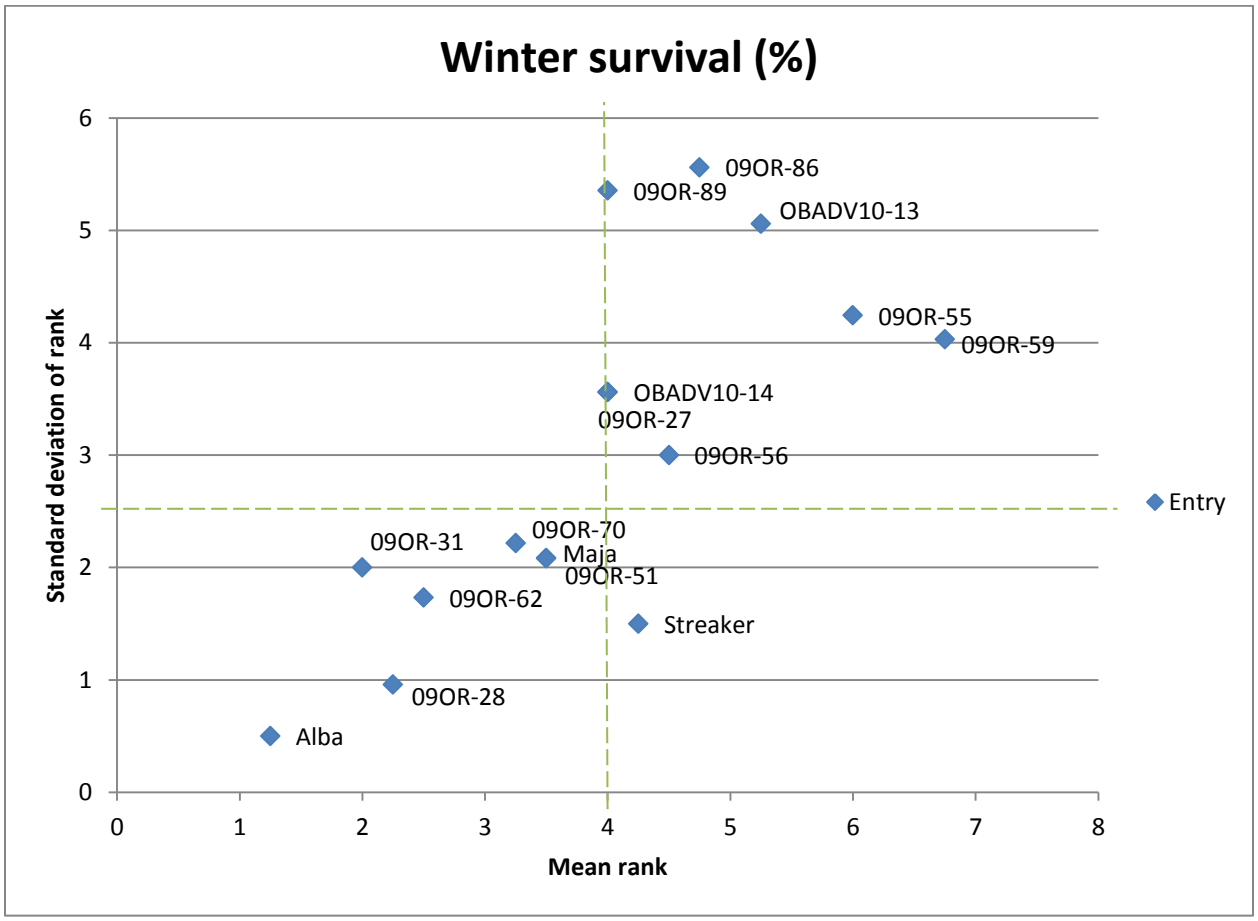


Figure 2.12. Pearson's simple correlation coefficients and *P* values for traits measured on all 16 entries in the OFOOD trial grown at Corvallis, OR; Pullman, WA; and Aberdeen, ID in 2012 and 2013.
*Significant at $P < 0.05$.
**Significant at $P < 0.01$.
‡ns, not significant.

	Trait						
Trait	**Protein**	**Kernel hardness**	**SRC-W**	**Yield**	**Test weight**	**Height**	**Heading date**
β-glucan	0.35**	0.40**	0.73**	-0.10ns‡	-0.42**	-0.23*	-0.19ns
Protein		0.04ns	0.50**	0.09ns	-0.07ns	-0.38**	0.30**
Kernel hardness			0.39**	0.08ns	-0.60**	0.24*	-0.30**
SRC-W				0.15ns	-0.53**	-0.25*	0.07ns
Yield					-0.17ns	0.06ns	0.22*
Test weight						0.11ns	0.15ns
Height							-0.32**

Registration of 'Streaker+' Barley

Brigid Meints, Alfonso Cuesta-Marcos, Scott Fisk, Andrew S. Ross, and Patrick M. Hayes.

Abstract

'Streaker+' (Reg. No._____) is a winter habit, hull-less, six-row barley (*Hordeum vulgare* L.) germplasm released by the Oregon Agricultural Experiment Station in 2014. It was named as such because it is a naked (hull-less) variety. Streaker+ consists of an equi-proportional blend of three experimental lines with the pedigree: Maja/Legacy, F1//Maja///Doyce. The three components of Streaker+ are all advanced lines from the Oregon State University Barley Breeding Program. Streaker+ is being released as a germplasm based on its novelty as a winter food barley with multi-colored grain that is adapted to the Pacific Northwest region. The rationale for the blend is that (i) the intended market - whole grain users of barley - prefer the multi-colored grain appearance and (ii) the mixture may show superior levels of resistance to biotic and abiotic stresses than the individual components. The "+" in Streaker+ differentiates this most recent germplasm release from the original Streaker blend that was tested and assessed for commercial potential.

Introduction

'Streaker+' (Reg. No._____) is a winter habit, hull-less, six-row barley (*Hordeum vulgare* L.) germplasm released by the Oregon Agricultural Experiment Station in 2014. It was named as such because it is a naked (hull-less) variety.

Streaker+ is an equi-proportional blend of three experimental lines with the pedigree: Maja/Legacy, F1//Maja///Doyce. The three components of Streaker+ are all advanced lines from the Oregon State University Barley Breeding Program and were tested with the experimental names: OR85, OR86, and OR911. In 2011 OR85, OR86, and OR911 were mixed in equal proportions and tested as 'Streaker' as a single entry in the OFOOD (Oregon Food Barley) trial. The "+" in Streaker+ designates the germplasm described in this release, which was generated by re-selection of Streaker in farmers' fields followed by head row purification (as described in the "Methods" section). The OFOOD trial was grown for two years (2011-12 and 2012-13) at eight locations, with five of the locations replicated over the two years for a total of 13 environments. Streaker+ is being released based on its novelty as a winter food barley with multi-colored grain (blue, brown, and white) that is adapted to the Pacific Northwest region of the U.S. The rationale for the blend is that (i) the intended market - whole grain users of barley - prefer the multi-colored grain appearance and (ii) the mixture may show superior levels of resistance to biotic and abiotic stresses than the individual components. All three components show varying degrees of susceptibility to scald (incited by *Rhynchosporium commune*), but all are resistant to barley stripe rust (incited by *Puccinia striiformis* f. sp. *hordei*), a disease that is prevalent throughout western Oregon and Washington. Streaker+ is susceptible to leaf rust (incited by *Puccinia hordei*). Mundt et al. (1994) showed that planting variety mixtures (even when the individual varieties show some disease susceptibility) can decrease disease severity. Therefore, Streaker+ may display

greater resistance to scald and leaf rust as a blend than as individual components. The principal end-use of Streaker+ is as grain for human consumption, but it can also be used as animal feed and may have some applications for malting and brewing.

The components of Streaker+ (OR85, OR86, and OR911) share the pedigree Maja/Legacy, F1//Maja///Doyce. 'Maja' is a six-row, hulled malting barley, with facultative growth habit, released by the Oregon Agricultural Experiment Station in 2006. 'Legacy' is a six-row, hulled malting barley, with spring growth habit, developed by Busch Agricultural Resources Inc. (http://anheuser-busch.com/). 'Doyce' is a six-row, hull-less feed barley, with winter growth habit, developed at Virginia Polytechnic Institute (Brooks *et al*., 2005) (see Fig. 3.1).

Methods

Generation Development and Line Selection

The cross between Maja/Legacy, F1//Maja and Doyce was made in 2003. Selections were made using a modified bulk-pedigree method. All generations from F_1 through F_4 head rows were grown under fall-planted conditions at the Oregon State University Hyslop Research Farm near Corvallis, OR. The F_2 populations were planted in bulk, from which individual heads were selected, threshed, bulked, and planted as an F_3 population. From the F_3 population, heads were selected and planted in F_4 head rows. Selected rows from the F_4 generation were harvested in bulk and advanced to a preliminary yield trial. Selections were subsequently grown in replicated, multi-environment yield trials in Oregon for multiple years. In 2008-09, two of the

Maja/Legacy, F1//Maja///Doyce crosses (F_5) were designated OR85 and OR86 and grown in the Oregon Barley Elite Trial (OBELT). This trial was grown again in 2009-10 and another Maja/Legacy, F1//Maja///Doyce cross (F_6) designated OR911 was included. In 2011 equal amounts of seed from OR85, OR86, and OR911 were combined into one entry called 'Streaker' in the OFOOD trial, which was grown for two years across eight locations in the Pacific Northwest.

Seed Increase and additional selection

During yield trial testing, there was also pre-commercial on-farm production. In the summer of 2013, 2100 heads of Streaker were selected from three local farmer's fields (700 per field). Field sizes ranged from 2 – 4 ha. The heads were threshed individually and only hull-less heads were selected. In the fall of 2013, 600 head rows from each farm were planted in separate blocks at the OSU Lewis Brown Research Farm near Corvallis, OR. Seed from each block, blended at equal weights, forms the Streaker+ germplasm.

Single head selections from each of the three components were used to grow a single plant for DNA extraction and genotyping using Single Nucleotide Polymorphisms (SNPs) under the auspices of the USDA-NIFA Triticeae Coordinated Agricultural Project (http://www.triticeaecap.org/), and these data are available at the T3 database (http://triticeaetoolbox.org/barley/). In the T3 database, the three components of Streaker+ can be found in the "LTT_TCFW6" panel. Based on the 6,895 molecular markers on the Infinium iSelect 9K genotyping chip, two of the three

components are 99.9% homozygous (OR85 and OR86) whereas OR911 is 92.7% homozygous. Pairwise genetic differences for the three components range from 12 to 20% (see Table 1). Therefore, the germplasm will capitalize on the heterogeneity present among the three genotypes, which are phenotypically very similar. All three components are similar in plant height and maturity and all have a soft kernel texture. Accordingly, the Streaker+ germplasm is sufficiently uniform for production and processing.

Statistical Analysis

All statistical analyses were conducted with Microsoft Excel (Microsoft Inc. Redmond, WA) and SAS for Windows version 9.3 (SAS Institute Inc. Cary, NC). Thirteen environments from the OFOOD trial were included in the comparison of Streaker and checks ‘Maja’ and ‘Alba’ (Graebner et al., submitted) in the analysis of agronomic and food quality traits, although not all traits were measured at all locations. Analysis of yield trial data was based on the trial means and was conducted both within and across locations. Mean separation tests were based on LSD (P = 0.05).

Characteristics

Botanical Description

Phenotypic selection resulted in a six-row barley germplasm in which all plants have semi-compact spikes and long rachilla hairs. There are rough and smooth awned

plants in the germplasm. OR86 is facultative, as measured by timely flowering under spring-sown conditions whereas OR85 and OR911 require vernalization to flower in an agronomically acceptable time frame. Under spring-planted conditions, OR86 flowered at 184 Julian days, comparable to 88Ab536 (facultative) which flowered at 182 Julian days and 'Full Pint' (spring) which flowered at 178 Julian days. OR85 and OR911 never flowered under spring-planted conditions. Under fall-sown conditions, the three components of Streaker+ flower within approximately one week of each other and are of similar plant height (data not shown). Grain color varies from blue, to brown, to white.

Agronomic Performance

Across all 13 environments, Streaker was lower yielding than Alba and Maja but the differences were not significant. Lower yield is expected from hull-less varieties as compared to hulled varieties due to the weight of the hull. Streaker was significantly shorter than Alba and similar in height to Maja. Grain from Streaker had significantly higher test weight than Alba and Maja under all growing conditions, as would be expected for hull-less vs. hulled varieties. Streaker flowered significantly earlier than Alba and was comparable to Maja under all growing conditions (Table 3.2). Pendleton, OR and Pullman, WA are classified as dryland locations because there is no irrigation applied and the annual rainfall averages are 420 mm $year^{-1}$ and 540 mm $year^{-1}$ respectively (Western Regional Climate Center). Under these conditions Streaker was significantly lower yielding than Alba and comparable to Maja. Streaker

was similar in height to Alba and Maja (Table 3.3). Corvallis, OR and Mount Vernon, WA are considered high-rainfall because the average rainfall is greater than 800 mm year^{-1} (Western Regional Climate Center). Under these conditions, Streaker yielded significantly less than Alba and similarly to Maja. Streaker was significantly shorter than Alba and comparable to Maja (Table 3.4). In Hermiston, OR and Aberdeen and Parma, ID supplemental irrigation is applied in accordance with local practice because the average annual rainfall is below 400 mm year^{-1}. Under irrigated conditions, Streaker did not have a significantly different yield from Alba or Maja. All varieties had similar plant heights (Table 3.5).

Disease Resistance

Disease was measured over two years under high rainfall conditions: no diseases were observed at the dryland or irrigated locations. Streaker was resistant to stripe rust at both high rainfall locations. The intensity of the stripe rust epidemics at these locations is apparent from the stripe rust severity of Thoroughbred, a susceptible line grown in an adjacent experiment, which averaged 82.5% severity over the two years. Streaker was significantly more susceptible to scald than Alba and comparable to Maja (Table 3.6). Seedling inoculation with five leaf rust isolates at the USDA-ARS Cereal Disease Laboratory revealed that Streaker was susceptible to all isolates. Leaf rust was observed under field conditions at Mount Vernon, WA for the first time in 2013; Streaker was rated as susceptible. Powdery Mildew (incited by *Blumeria graminis* f. sp. *hordei*) is occasionally observed in the Pacific Northwest of the US.

Intense epidemics occurred at Mount Vernon, WA in 2012 and 2013 and in Corvallis, OR in 2014. In Mount Vernon in 2012 and 2013, no mildew was observed on Streaker, but adjacent plots of the variety Full Pint were rated up to 53% leaf coverage. In Corvallis 2014, Streaker had 10% mildew severity, Alba had 30%, and Maja had 20%.

Food Quality

Streaker had higher protein than Alba and Maja under all growing conditions Maja (Table 3.2). Streaker had similar levels of grain β-glucan (AACC-International Method 32-23.01) to Alba and Maja under all growing conditions Maja (Table 3.2). Streaker had a lower solvent retention capacity (AACC-International Approved Method 56-11.02) for water than Alba and Maja under high rainfall and irrigated conditions (Tables 3.4 and 3.5) and a similar solvent retention capacity for water under dryland conditions (Table 3.3). Under all growing conditions, Streaker had significantly softer kernels than Alba and similar kernel hardness to Maja (Table 3.2). Additional data on other nutritional traits, processing characteristics, and food products are available under the heading 'Food Barley Standard Reference Panel' at http://barleyworld.org/research-highlights. In 2006, the US-FDA approved a health claim for barley that states: "foods containing barley to claim that they reduce the risk of coronary heart disease. Specifically, whole grain barley and dry milled barley products such as flakes, grits, flour, and pearled barley, which provide at least 0.75 grams of soluble fiber per serving" (21 CFR 101.81) (Ames and Rhymer, 2008;

National Barley Foods Council, 2003). Based on the average β-glucan content of Streaker+, in order to receive the daily recommended soluble fiber, a person would have to eat at least 17g of steamed grain or 44g of bread made with 40% barley flour. This amounts to a small side dish of steamed grain or only two slices of bread per serving.

Winter-hardiness

Differential winter survival was observed in four of the thirteen environments. In these environments (Pullman, WA; Aberdeen and Parma, ID), the low temperature tolerance of Streaker was not significantly different than Maja and significantly lower than Alba. The high survival values in these field trials indicate that Streaker has a level of low temperature tolerance at least comparable to that of the checks (Table 3.6).

Availability

Seed from the 2014 germplasm production is maintained by the Barley Project at Oregon State University, Corvallis, OR 97331. Seed for research purposes will be available on request from the corresponding author for at least 5 years. It is requested that an appropriate recognition of source be given when any component of the Streaker+ blend contributes to the development of new germplasm or varieties. Grain for human consumption is available through Camas Country Mill in Junction City, OR (camas.squarespace.com).

Acknowledgements

The development of Streaker+ is supported by the Oregon Agricultural Experiment Station, the Oregon Wheat Commission, the Idaho Barley Commission, Formula Grant no. 2013-31100-06041from the USDA National Institute of Food and Agriculture, and USDA-NIFA TCAP Project No. 2011-68002-30029. Thanks to all the cooperators who have participated in collaborative testing of the OFOOD trial, including Dr. Juliet Marshall and Chad Jackson (University of Idaho; Aberdeen, ID), Mr. Mathias Kolding (Oregon State University emeritus; Hermiston, OR), Dr. Kevin Murphy (Washington State University; Pullman, WA), Brook Brouwer and Dr. Steve Jones (Washington State University; Mount Vernon, WA), Dr. Bradford Brown (University of Idaho; Parma, ID), and Dr. Steven Petrie and Karl Rhinhart (Oregon State University; Pendleton, OR). Thanks to members of the OSU Barley Project for their assistance with food quality trait measurement. Special thanks to Tom Hunton (Junction City, OR), Scott Sayer (Brownsville, OR), and David Simmons (Salem, OR) for growing and promoting Streaker+.

References

Ames NP and Rhymer CR (2008) Issues surrounding health claims for barley. J Nutr 138: 1237S-1243S.

Brooks, W. S., Griffey, C. A., Price, A. M., Pridgen, T. H., & Rohrer, W. L. (2005). Registration of 'Doyce' Hulless Barley the ventral furrow. *Crop science*, *45*, 791-792.

Mundt, C. C., Hayes, P. M., & Schön, C. C. (1994). Influence of barley variety mixtures on severity of scald and net blotch and on yield. *Plant pathology*, *43*(2), 356-361.

National Barley Foods Council (2003) Petition for unqualified health claim: Barley β-glucan soluble fiber and barley products containing β-glucan soluble fiber and coronary heart disease. Submitted to the FDA on September 25, 2003.

Western Regional Climate Center. SOD USA Climate Archive. Available at http://www.wrcc.dri.edu/summary/ (verified Apr. 23, 2014).

Figure 3.1. Pedigree contributing to Streaker+. Varieties in bold were developed by other breeding programs. Underlined varieties were released from the Oregon Agricultural Experiment Station with the year of release in parentheses.

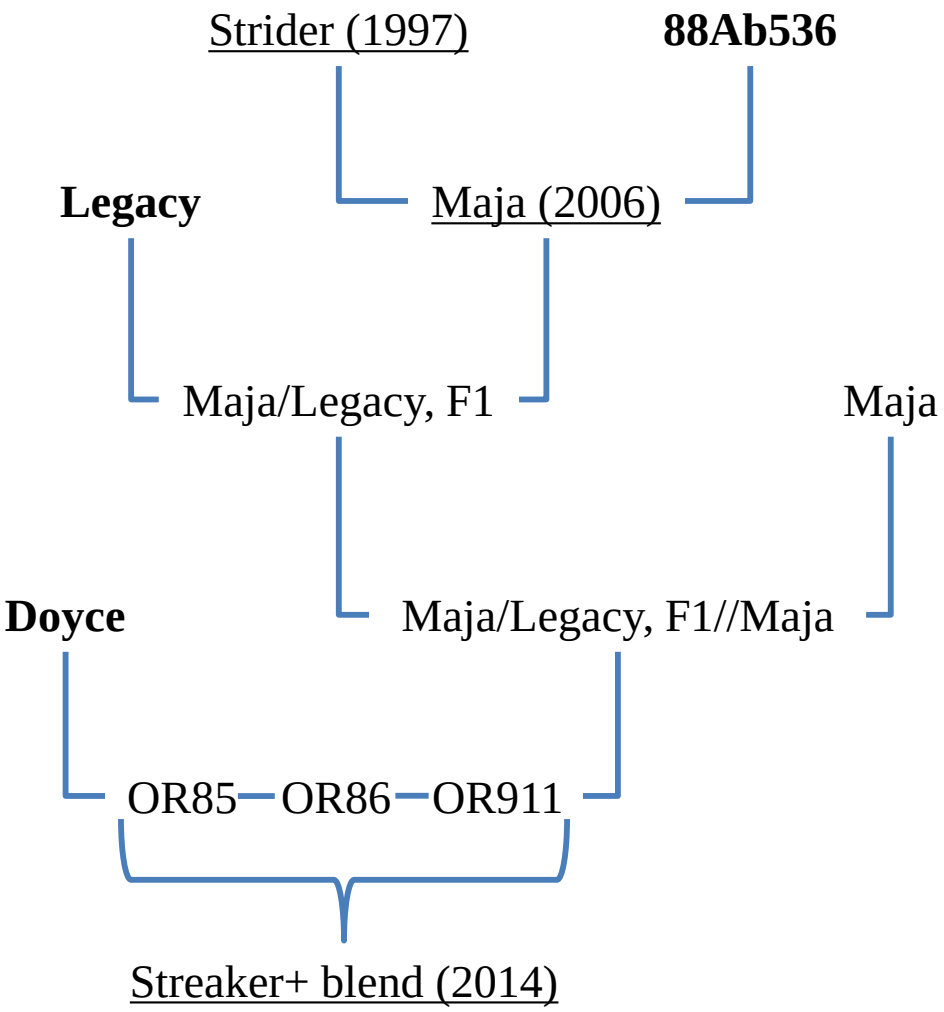


Table 3.1. Pairwise genetic differences between each of the three components of Streaker+ based on 6,895 molecular markers on the Infinium iSelect 9K genotyping chip.

	OR85	OR86	OR911
OR85	--	18%	20%
OR86		--	12%
OR911			--

Table 3.2. Agronomic performance and food quality of Streaker and check varieties across 13 environments (4 high rainfall, 4 dryland, 5 irrigated).*
‡ Same letter indicates no significant difference between lines.
*Corvallis, Hermiston, Lewis-Brown, and Pendleton, OR; Mount Vernon and Pullman, WA; Aberdeen and Parma, ID.

	Agronomic traits				Food quality traits			
	Yield	Heading date	Plant Height	Test weight	β-glucan	Protein	Solvent Retention Capacity (water)	Kernel Hardness
	kg ha^{-1}	Julian days	cm	kg hL^{-1}	% (w/w)	%	%	SKCS units
Streaker	6238a‡	134b	90.7b	74.3a	4.1ab	12.2a	100.8b	46.1c
Alba	7299a	142a	99.3a	65.9b	4.3a	11.0b	107.4a	69.1a
Maja	6746a	136b	90.6b	65.0b	3.9b	11.2b	100.5b	52.4b
# env.	9	7	13	9	11	11	10	13
LSD (*P* = 0.05)	1278	4	4.9	2.8	0.3	0.7	6.1	5.5

Table 3.3. Agronomic performance and food quality of Streaker and check varieties across 4 dryland environments.*
‡ Same letter indicates no significant difference between lines.
* Pendleton, OR and Pullman, WA.

	Agronomic traits				Food quality traits			
	Yield	Heading date	Plant Height	Test weight	β-glucan	Protein	Solvent Retention Capacity (water)	Kernel Hardness
	kg ha^{-1}	Julian days	cm	kg hL^{-1}	% (w/w)	%	%	SKCS units
Streaker	5860b‡	144c	89.6a	74.6a	4.3a	14.2a	108.2a	45.5b
Alba	7512a	147a	95.6a	66.8b	4.3a	12.2b	111.6a	70.4a
Maja	6023b	146b	87.0a	68.1b	4.0b	12.7b	99.7a	48.8b
# env	3	2	4	3	4	4	4	4
LSD (*P* = 0.05)	1172	0	11.4	2.0	0.3	1.5	12.0	5.7

Table 3.4. Agronomic performance and food quality of Streaker and check varieties across 4 high rainfall environments.*
‡ Same letter indicates no significant difference between lines.
* Corvallis and Lewis-Brown, OR; Mount Vernon, WA.

	Agronomic traits				Food quality traits			
	Yield	Heading date	Plant Height	Test weight	β-glucan	Protein	Solvent Retention Capacity (water)	Kernel Hardness
	kg ha^{-1}	Julian days	cm	kg hL^{-1}	% (w/w)	%	%	SKCS units
Streaker	4635b‡	123b	92.8b	75.7a	4.4a	12.0a	94.2a	50.6b
Alba	8243a	136a	111.0a	67.0b	4.2a	9.7b	104.5a	74.9a
Maja	4927b	125b	96.2b	62.4b	3.7a	10.7ab	100.9a	56.9b
# env	2	3	4	3	3	3	2	4
LSD (*P* = 0.05)	2163	4	5.1	8.5	0.8	1.6	17.9	11.0

Table 3.5. Agronomic performance and food quality of Streaker and check varieties across 5 irrigated environments.*
‡ Same letter indicates no significant difference between lines.
* Hermiston, OR; Aberdeen and Parma, ID.

	Agronomic traits				Food quality traits			
	Yield	Heading date	Plant Height	Test weight	β-glucan	Protein	Solvent Retention Capacity (water)	Kernel Hardness
	kg ha^{-1}	Julian days	cm	kg hL^{-1}	% (w/w)	%	%	SKCS units
Streaker	7323ab‡	141b	89.9a	73.0a	3.9a	10.8a	96.8a	43.0b
Alba	6667b	145a	93.0a	64.7b	4.3a	10.4a	104.5a	63.4a
Maja	8198a	143ab	89.0a	64.8b	3.9a	10.1a	101.1a	51.7ab
# env	4	2	5	4	5	5	4	5
LSD (*P* = 0.05)	1398	4	8.4	3.0	0.6	1.2	9.9	13.7

Table 3.6. Reaction of Streaker and check varieties to barley stripe rust and scald, and winter survival.
* Based on a 1-9 rating scale where 1 = most resistant and 9 = most susceptible.

	Barley stripe rust	**Scald**	**Winter Survival**
	%	1-9 scale*	%
Streaker	6.7	7.1	92.0
Alba	3.3	0.9	97.9
Maja	0.0	6.8	93.0
# env	3	4	5
LSD (P = 0.05)	13.1	2.7	4.2

General Conclusions

As discussed in chapter one, this research contributes to the larger food barley improvement effort. Despite an overall decline in food barley consumption over the last few centuries, food barley germplasm development and quality characterization is making a comeback in many areas of the world. As consumers realize the nutrition and taste benefits of barley, commercial production increases and there becomes a need for new varieties adapted to a number of different regions. Oregon State University is on the forefront of food barley research, thanks to extensive regional, national, and international collaboration.

Our results from the OFOOD experiment (chapter two) will help to meet needs of farmers, consumers, and processors. The overall grain yields achieved with this germplasm are much higher than those reported for spring barley germplasm by Rey et al. (2009). Additionally, despite some evidence that the hull-less trait is associated with lower yield and vigor, there were no significant differences between the hulled and hull-less classes in this germplasm. Therefore, our future food projects will be focused exclusively on breeding hull-less lines. This is due to an interest in the whole grain benefits and the processing difficulties that arise with pearling. We found that waxy starch played an important role in determining quality traits, including β-glucan, kernel hardness, and SRC-W. Holtekjolen et al. (2008) reports that waxy starch can lead to difficulties in the baking process, if the appropriate amount of water is not used. Therefore, our efforts now focus on hull-less non-waxy types, with

modest β-glucan levels. Streaker+ will be released in 2014, and 09OR-86 is a candidate for release.

In chapter three, we described the novel approach to releasing Streaker+ as a germplasm. We believe that due to the mixture of genotypes, Streaker+ will show greater disease resistance, and the blend of colors and nutritional qualities will fill a niche market. A number of farmers in the Willamette Valley have expressed interest in Streaker and approximately 30 acres were grown in the 2012-13 year. The harvested grain has been cracked to allow better cooking time, milled to flour for baking, and 23 tons have been rolled to create barley flakes. A local school district has altered a recipe for breakfast bars to accommodate the barley flakes and has had a great response from students. A popped barley snack, trail mix, and granola bar are in development. With new and exciting germplasm just down the pipeline, we hope to invigorate our local barley market by engaging farmers, processors, and consumers and to assist in developing markets worldwide.

Cereal grains, which carry many essential and nutritional components, will be critical to the improvement of human diets in an effort to alleviate the many ills associated with the sedentary lifestyle of much of the developed world (Lafiandra et al., 2014). Much progress has been made over the last decade in food barley breeding and characterization in different breeding programs around the world; our program will continue to focus on breeding nutritious and delicious barley varieties with good food quality that are adapted for the Pacific Northwest.

Bibliography

AACC International. Approved Methods of Analysis, 11th Ed. Method 56-11.02. Solvent Retention Capacity Profile. Approved June 3, 2009. AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-56-11.02

AACC International. Approved Methods of Analysis, 11th Ed. Method 32-23.01. β-glucan Content of Barley and Oats- Rapid Enzymatic Procedure. Approved November 3, 1999. AACC International, St. Paul, MN, U.S.A. http://dx.doi.org/10.1094/AACCIntMethod-32-23.01

Abdel-Aal ESM, Choo TM, Dhillon S, Rabalski I (2012) Free and bound phenolic acids and total phenolics in black, blue, and yellow barley and their contribution to free radical scavenging capacity. Cereal Chem. 89(4): 198-204

Abraha A, Uhlen AK, Abay F, Sahlstrøm S, Bjørnstad Å (2013) Genetic variation in barley enables a high quality, the Ethiopian staple flat bread, comparable to Tef. Crop Sci. 53(5): 2040-2050

AbuMweis SS, Jew S, Ames NP (2010) β-glucan from barley and its lipid-lowering capacity: a meta-analysis of randomized, controlled trials. Eur J Clin Nutr 64: 1472-1480

Ames NP and Rhymer CR (2008) Issues surrounding health claims for barley. J Nutr 138: 1237S-1243S

Ames N, Rhymer C, Rossnagel B, Therrien M, Ryland D, Dua S, Ross K (2006) Utilization of diverse hulless barley properties to maximize food product quality. Cereal Food World. 51(1): 23-28

Anker-Nilssen K, Sahlstrøm S, Knutsen SH, Holtekjølen AK, Uhlen AK (2008) Influence of growth temperature on content, viscosity and relative molecular weight of water-soluble β-glucans in barley (Hordeum vulgare L.) J Cereal Sci. 48(3): 670-677

Arndt EA (2006) Whole-grain barley for today's health and wellness needs. Cereal Food World. 51: 20-22

Baik BK and Czuchajowska Z (1997) Barley in udon noodles. Food Sci Technol Int. 3: 423-435

Baik BK, Newman CW, Newman RK (2011) Food uses of barley. In: Ullrich SE (eds) Barley: Production, Improvement, and Uses. Wiley-Blackwell, 532-562

Baik BK and Ullrich SE (2008) Barley for food: characteristics, improvement, and renewed interest. J Cereal Sci. 48: 233-242

Bays H, Frestedt JL, Bell M, Williams C, Kolberg L, Schmelzer W, Anderson JW (2011) Reduced viscosity barley β-glucan versus placebo: a randomized controlled trial of the effects on insulin sensitivity for individuals at risk for diabetes mellitus. Nutr Metab. 8: 10

Beecher B, Bowman J, Martin JM, Bettge AD, Morris CF, Blake TK, Giroux MJ (2002) Hordoindolines are associated with a major endosperm- texture QTL in barley (Hordeum vulgare). Genome. 45(3): 584-591

Behall KM, Scholfield DJ, Hallfrisch JG (2006) Barley β-glucan reduces plasma glucose and insulin responses compared with resistant starch in men. Nutr Res. 26: 644-650

Behall KM, Scholfield DJ, Hallfrisch JG (2004a) Diets containing barley significantly reduce lipids in mildly hypercholesterolemic men and women. Am J Clin Nutr. 80(5): 1185-1193

Behall KM, Scholfield DJ, Hallfrisch JG (2004b) Lipids significantly reduced by diets containing barley in moderately hypercholesterolemic men. J Am Coll Nutr. 23: 55-62

Bellido GG and B T (2009) Anthocyanin composition and oxygen radical scavenging capacity (ORAC) of milled and pearled purple, black, and common barley. J Agr Food Chem. 57(3): 1022-1028

Bendelow VM (1975) Determination of non-starch polysaccharides in barley breeding programmes. J I Brewing. 81(2): 127-130

Bernardo R (2008) Molecular markers and selection for complex traits in plants: learning from the last 20 years. Crop Sci. 48: 1649–1664

Bhatty RS (1997) Milling of Regular and Waxy Starch Hull-less Barleys for the Production of Bran and Flour 1. Cereal Chem. 74(6): 693-699

Bhatty RS (1999) The potential of hull-less barley. Cereal Chem. 76(5): 589-599

Bhatty RS and Rossnagel BG (1998) Comparison of pearled and unpearled Canadian and Japanese barleys. Cereal Chem. 75(1): 15-21

Biorklund M, van Rees A, Mensink RP, Onning G (2005) Changes in serum lipids and postprandial glucose and insulin concentrations after consumption of beverages with β-glucans from oats or barley: a randomised dose-controlled trial. Eur J Clin Nutr. 59: 1272–1281

Brennan CS and Cleary LJ (2005) The potential use of cereal (1→ 3, 1→ 4)-β-D-glucans as functional food ingredients. J Cereal Sci. 42(1): 1-13

Brinch-Pedersen H, Borg S, Tauris B, Holm PB (2007) Molecular genetic approaches to increasing mineral availability and vitamin content of cereals. J Cereal Sci. 46(3): 308-326

Brooks WS, Griffey CA, Price AM, Pridgen TH, Rohrer WL (2005) Registration of 'Doyce' Hulless Barley the ventral furrow. Crop Sci. 45: 791-792

Brouns F, Kettlitz B, Arrigoni E (2002) Resistant starch and "the butyrate revolution". Trends Food Sci Tech. 13(8): 251-261

Brown AHD, Burdon JJ, Garvin DF, Abbott DC, Read BJ (1996) The effect of combining scald resistance genes on disease levels, yield and quality traits in barley. Theor Appl Genet. 93(3): 361-366

Brueggeman R, Steffenson BJ, Kleinhofs A (2009) The rpg4/Rpg5 stem rust resistance locus in barley. Cell Cycle. 8(7): 977-981

Cai S, Yu G, Chen X, Huang Y, Jiang X, Zhang G, Jin X (2013) Grain protein content variation and its association analysis in barley. BMC Plant Biol. 13(1): 35

Capo-chichi L, Kenward K, Nyachito J, Anyia A (2012) Nud locus and the effects on seedling vigour related traits for genetic improvement of hulless barley. Journal of Plant Science and Molecular Breeding. 1(1): 2

Cascadia Grains Conference 2013 http://cascadiagrains.com/events-registration/past-events/2013-conference/ accessed 2014-02-12

Cascadia Grains Conference 2014 http://cascadiagrains.com/events-registration/past-events/2014-conference/ accessed 2014-02-12

Casiraghi MC, Garsetti M, Testolin G, Brighenti F (2006) Post-prandial responses to cereal products enriched with barley β-glucan. J Am Coll Nutr. 25: 313-320

Champ M (2013) Metabolic Effects of Resistant Starch. In: Shi YC and Maningat CC (eds.) Resistant Starch: Sources, Applications and Health Benefits. John Wiley & Sons, 2013 p. 229-249

Cho SS, Prosky L, Dreher M (1999) Complex carbohydrates in food. Marcel Dekker, New York.

Choo TM, Ho KM, Martin RA (2001) Genetic analysis of a hulless × covered cross of barley using doubled-haploid lines. Crop Sci. 41(4): 1021-1026

Chutimanitsakun Y, Cuesta-Marcos A, Chao S, Corey A, Filichkin T, Fisk S, Kolding M, Meints B, Ong YL, Rey JI, Ross AS, Hayes PM (2013) Application of marker-assisted selection and genome-wide association scanning to the development of winter food barley germplasm resources. Plant Breeding. 132(6): 563-570

Cistué L, Cuesta-Marcos A, Chao S, Echávarri B, Chutimanitsakun Y, Corey A, Filichkin T, Garcia-Mariño N, Romagosa I, Hayes P (2011) Comparative mapping of the Oregon Wolfe Barley using doubled haploid lines derived from female and male gametes. Theor Appl Genet. 122: 1399-1410

Crosbie GB and Ross AS (2004) Asian wheat flour noodles. In: Wrigley CW, Corke H, Walker CE (eds.) The Encyclopedia of Grain Science, Vol 2. Elsevier Academic Press, Oxford UK, 304-312

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

Dai F, Nevo E, Wu D, Comadran J, Zhou M, Qiu L, Chen Z, Beiles A, Chen G, Zhang G (2012) Tibet is one of the centers of domestication of cultivated barley. P Natl Acad Sci USA. 109(42): 16969-16973

Di Cagno R, De Angelis M, Lavermicocca P, De Vincenzi M, Giovannini C, Faccia M, Gobbetti M (2002) Proteolysis by sourdough lactic acid bacteria: effects on wheat flour protein fractions and gliadin peptides involved in human cereal intolerance. Appl Environ Microb. 68(2): 623-633

Dickin E, Steele K, Edwards-Jones G, Wright D (2012) Agronomic diversity of naked barley (Hordeum vulgare L.): a potential resource for breeding new food barley for Europe. Euphytica. 184(1): 85-99

Edney MJ, Rossnagel BG, Endo Y, Ozawa S, Brophy M (2002) Pearling quality of Canadian barley varieties and their potential use as rice extenders. J Cereal Sci. 36: 295-305

FAOSTAT (2011) Production and trade. (http://faostat.fao.org/site/339/default.aspx) Verified February 13, 2013. FAO, Rome, Italy

Fardet A (2010) New hypotheses for the health-protective mechanisms of whole-grain cereals: what is beyond fibre? Nutr Res Rev. 23.01: 65-134

Fardet A (2013) Whole Grains from a Mechanistic View. Proceedings of the Whole Grains Summit 2012. T. Schryver Ed. http://www.aaccnet.org/publications/plexus/cfwplexus/library/books/Documents/WholeGrainsSummit2012/CPLEX-2013-1001-01B.pdf accessed 2014-02-14

Fardet A and Rock E (2013) The search for a new paradigm to study micronutrient and phytochemical bioavailability: From reductionism to holism. Med Hypotheses. http://dx.doi.org/10.1016/j.mehy.2013.11.035

Fastnaught CE (2010) Barley fiber. In: Cho SS and Samuel P (eds.) Fiber Ingredients: Food Applications and Health Benefits. CRC Press, 323-358

Fastnaught CE, Berglund PT, Holm ET, Fox GJ (1996) Genetic and environmental variation in β-glucan content and quality parameters of barley for food. Crop Sci. 36(4): 941-946

Fisk SP, Cuesta-Marcos A, Cistue L, Russell J, Smith K, Baenziger S, Bedo Z, Corey A, Filichkin T, Karsai I, Waugh R, Hayes PM (2013) FR-H3: a new QTL to assist in the development of fall-sown barley with superior low temperature tolerance. Theor Appl Genet. 126: 335-347

Frølich W (1990) Chelating properties of dietary fiber and phytate. The role for mineral availability. In: New Developments in Dietary Fiber. Springer US, 83-93

Fung KY, Cosgrove L, Lockett T, Head R, Topping DL (2012) A review of the potential mechanisms for the lowering of colorectal oncogenesis by butyrate. Brit J Nutr. 1(1): 1-12

Gamlath J, Aldred GP, Panozzo JF (2008) Barley (1→ 3; 1→ 4)-β-glucan and arabinoxylan content are related to kernel hardness and water uptake. J Cereal Sci. 47(2): 365-371

Gänzle MG, Loponen J, Gobbetti M (2008) Proteolysis in sourdough fermentations: mechanisms and potential for improved bread quality. Trends Food Sci Tech. 19(10): 513-521

Garvin DF, Brown AHD, Burdon JJ (1997) Inheritance and chromosome locations of scald-resistance genes derived from Iranian and Turkish wild barleys. Theor Appl Genet. 94(8): 1086-1091

Gauch HG (1988) Model selection and validation for yield trials with interaction. Biometrics. 44: 705–715

Go AS, Mozaffarian D, Roger VL, Benjamin EJ, Berry JD, Borden WB, ... Turner MB (2013) on behalf of the American Heart Association Statistics Committee and Stroke Statistics Subcommittee. Heart disease and stroke statistics—2013 update: a report from the American Heart Association. Circulation. 127(1): e1-e240

Gollob HF (1968) A statistical model which combines features of factor analytic and analysis of variance techniques. Psychometrika. 33: 73–115

Gong LX, Jin C, Wu LJ, Wu XQ, Zhang Y (2012) Tiβn Hull-less Barley (Hordeum vulgare L.) as a potential source of antioxidants. Cereal Chem. 89(6): 290-295

Grando S and Macpherson HG (2005) (eds.) Food Barley: Importance, Uses and Local Knowledge. Proceedings of the International Workshop on Food Barley Improvement, 14-17 January 2002, Hammamet, Tunisia. ICARDA, Aleppo, Syria, x+156 pp. En.

Gray D, Abdel-Aal ESM, Seetharaman K, Kakuda Y (2010) Differences in viscosity and textural properties of selected barley cultivars as influenced by pearling and cooking. Food Chem. 120: 402-409

Gupta M, Bawa AS, Semwal AD (2008) Effect of barley flour incorporation on the instrumental texture of sponge cake. Int J Food Pro. 12: 243-251

Guttieri MJ and Souza E (2003) Sources of variation in the solvent retention capacity test of wheat flour. Crop Sci. 43(5): 1628-1633

Hatcher DW, Lagasse S, Dexter JE, Rossnagel B, Izydorczyk M (2005) Quality characteristics of yellow alkaline noodles enriched with hull-less barley flour. Cereal Chem. 82: 60-69

Heffner E, Jannink JL, Sorrells ME (2011) Genomic selection accuracy using multifamily prediction models in a wheat breeding program. Plant Gen. 4: 65–75

Heffner EL, Lorenz AJ, Jannink JL, Sorrells ME (2010) Plant breeding with genomic selection: gain per unit time and cost. Crop Sci. 50: 1681–1690

Heslot N, Yang HP, Sorrells ME, Jannink JL (2012) Genomic selection in plant breeding: a comparison of models. Crop Sci. 52: 146-160

Hinata M, Ono M, Midorikawa S, Nakanishi K (2007) Metabolic improvement of male prisoners with type 2 diabetes in Fukushima Prison, Japan. Diabetes Res Clin Pr. 77(2): 327-332

Holtekjølen AK, Olsen HHR, Færgestad EM, Uhlen AK, and Knutsen SH (2008) Variations in water absorption capacity and baking performance of barley varieties with different polysaccharide content and composition. Food Sci Technol-LEB (10): 2085-2091

Holtekjølen AK, Sahlstrom S, Knutsen SH (2011) Phenolic contents and antioxidant activities in covered whole-grain flours of Norwegian barley varieties and in fractions obtained after pearling. Acta Agric Scand. 61: 67-74

Hu G and Burton C (2008) Modification of standard enzymatic protocol to a cost-efficient format for mixed-linkage (1→3,1→4)-β-d-glucan measurement. Cereal Chem. 85: 648-653

Islamovic E, Obert DE, Oliver RE, Harrison SA, Ibrahim A, Marshall JM, Miclaus KJ, Hu G, Jackson EW (2013) Genetic dissection of grain β-glucan and amylose content in barley (*Hordeum vulgare* L.). Mol Breeding. 31: 15-25

Iwata H and Jannink JL (2011) Accuracy of genomic selection prediction in barley breeding programs: A simulation study based on the real single nucleotide polymorphism data of barley breeding lines. Crop Sci. 51: 1915–1927

Izydorczyk MS (2010) Variations in content and molecular structure of barley nonstarch polysaccharides associated with genotypic and cellular origin. Cereal Chem. 87: 376-384

Izydorczyk MS, Chornick TL, Paulley FG, Edwards NM, Dexter JE (2008) Physicochemical properties of hull-less barley fibre-rich fractions varying in particle size and their potential as functional ingredients in two-layer flat bread. Food Chem. 108: 561-570

Izydorczyk MS and Dexter JE (2008) Barley β-glucans and arabinoxylans: molecular structure, physicochemical properties, and uses in food products–a review. Food Res Int. 41(9): 850-868

Izydorczyk MS, Lagasse S, Hatcher DW, Dexter JE, Rossnagel BG (2005) The enrichment of Asian noodles with fiber-rich fractions derived from roller milling of hull-less barley. J Sci Food Agr. 85: 2094-2104

Izydorczyk MS, Storsley J, Labossiere D, MacGregor AW, Rossnagel BG (2000) Variation in total and soluble β-glucan content in hulless barley: effects of thermal, physical, and enzymic treatments. J Agr Food Chem. 48(4): 982-989

Jones JM (2010) Dietary Fibre‘s Co-Passengers: Is It the Fibre or the Co-Passengers pp 365-378. In:. Van Der Kamp J.W., Jones J, Mccleary B, Topping DJ (eds.) Dietary Fibre: New Frontiers for Food and Health. Wageningen Academic Publishers: Wageningen, The Netherlands

Jones JM, Reicks M, Adams J, Fulcher G, Weaver G, Kanter M, Marquart L (2002) The importance of promoting a whole grain foods message. J Am Coll Nutr. 21(4):293-297

Keenan JM, Goulson M, Shamliyan T, Knutson N, Kolberg L, Curry L (2007) The effects of concentrated barley β-glucan on blood lipids in a population of hypercholesterolaemic men and women. Brit J Nutr. 97(06): 1162-1168

Keogh GF, Cooper GJ, Mulvey TB, McArdle BH, Coles GD, Monro JA, Poppitt SD (2003) Randomized controlled crossover study of the effect of a highly β-glucan–enriched barley on cardiovascular disease risk factors in mildly hypercholesterolemic men. Am J Clin Nutr. 78(4): 711-718

Kim H, Stote KS, Behall KM, Spears K, Vinyard B, Conway JM (2009) Glucose and insulin responses to whole grain breakfasts varying in soluble fiber, β-glucan. Eur J Nutr. 48: 170-175

Kim H, Turowski M, Anderson WK, Young SA, Kim Y, Yokoyama W (2011a) Supplementation of hydroxypropyl methylcellulose into yeast leavened all-whole grain barley bread potentiates cholesterol-lowering effect. J Agr Food Chem. 59(14): 7672-7678

Kim MJ, Hyun JN, Kim JA, Park JC, Kim MY, Kim JG, Lee SJ, Chun SC, Chung IM (2007) Relationship between phenolic compounds, anthocyanins content and antioxidant activity in colored barley germplasm. J Agr Food Chem. 55(12): 4802-4809

Kim Y and Yokoyama WH (2011b) Physical and sensory properties of all-barley and all-oat breads with additional Hydroxypropyl Methylcellulose (HPMC) β-glucan. J Agr Food Chem. 59: 741-746

King RA, Noakes M, Bird AR, Morell MK, Topping DL (2008) An extruded breakfast cereal made from a high amylose barley cultivar has a low glycemic index and lower plasma insulin response than one made from a standard barley. J Cereal Sci. 48: 526-530

Kinner M, Nitschko S, Sommeregger J, Petrasch A, Linsberger-Martin G, Grausgruber H, Berghofer E, Siebenhandl-Ehn S (2011) Naked barley—optimized recipe for pure barley bread with sufficient β-glucan according to the EFSA health claims. J Cereal Sci. 53: 225-230

Kweon M, Slade L, Levine H (2011) Solvent Retention Capacity (SRC) Testing of Wheat Flour: Principles and value in predicting flour functionality in different wheat-based food processes and in wheat breeding-A review. Cereal Chem. 88(6): 537-552

Lafiandra D, Riccardi G, and Shewry PR (2014) Improving Cereal Grain Carbohydrates for Diet and Health. J Cereal Sci.

Lagassé SL, Hatcher DW, Dexter JE, Rossnagel BG, Izydorczyk MS (2006) Quality characteristics of fresh and dried white salted noodles enriched with flour from hull-less barley genotypes of diverse amylose content. Cereal Chem. 83: 202-210

Li H, Singh RP, Braun HJ, Pfeiffer WH, Wang J (2013) Doubled haploids versus conventional breeding in CIMMYT wheat breeding programs. Crop Sci. 52: 74–83

Li J, Kaneko T, Qin LQ, Wang J, Wang Y (2003) Effects of barley intake on glucose tolerance, lipid metabolism, and bowel function in women. Nutrition. 19(11): 926-929

Lorenz AJ, Smith KP, Jannink JL (2012) Potential and optimization of genomic selection for Fusarium head blight resistance in six-row barley. Crop Sci. 52: 1609-1621

Lorenzana RE and Bernardo R (2009) Accuracy of genotypic value predictions for marker-based selection in bi-parental plant populations. Theor Appl Genet. 120: 151–161

Meuwissen TH, Hayes BJ, Goddard ME (2001) Prediction of total genetic value using genome-wide dense marker maps. Genetics. 157: 1819–1829

Morrell PL and Clegg MT (2007) Genetic evidence for a second domestication of barley (Hordeum vulgare) east of the Fertile Crescent. P Natl Acad Sci USA. 104(9): 3289-3294

Mundt CC, Hayes PM, Schön CC (1994) Influence of barley variety mixtures on severity of scald and net blotch and on yield. Plant Pathol. 43(2): 356-361

Murovec J and Bohanec B (2011) Haploids and doubled haploids in plant breeding. In: Plant breeding. I. Abdurakhmonov (ed.) InTech

Murphy K, Lammer D, Lyon S, Carter B, Jones SS (2005) Breeding for organic and low-input farming systems: An evolutionary–participatory breeding method for inbred cereal grains. Renew Agr Food Syst. 20(01): 48-55

Nair S, Ullrich SE, Blake TK, Cooper B, Griffey CA, Hayes PM, Hole DJ, Horsley RD, Obert DE, Smith KP, Muehlbauer GJ, Baik, B-K (2010) Variation in kernel hardness and associated traits in US barley breeding lines. Cereal Chem. 87: 461-466

Nair S, Knoblauch M, Ullrich S, Baik BK (2011a) Microstructure of hard and soft kernels of barley. J Cereal Sci. 54(3): 354-362

Nair S, Ullrich SE, Baik BK (2011b) Association of barley kernel hardness with physical grain traits and food processing parameters. Cereal Chem. 88(2): 147-152

National Barley Foods Council (2003) Petition for unqualified health claim: Barley β-glucan soluble fiber and barley products containing β-glucan soluble fiber and coronary heart disease. Submitted to the FDA on September 25, 2003.

Nevo E and Shewry PR (1992) Origin, evolution, population genetics and resources for breeding of wild barley, Hordeum spontaneum, in the Fertile Crescent. Barley: genetics, biochemistry, molecular biology and biotechnology. 19-43

Newman RK and Newman CW (2008) Barley for food and health: Science, technology, and products. John Wiley & Sons.

Newton AC, Flavell AJ, George TS, Leat P, Mullholland B, Ramsay L, Bingham IJ (2011) Crops that feed the world 4. Barley: a resilient crop? Strengths and weaknesses in the context of food security. Food Security. 3(2): 141-178

Nugent AP (2005) Health properties of resistant starch. British Nutrition Foundation, Nutrition Bulletin. 30: 27–54

Park Y, Subar AF, Hollenbeck A, Schatzkin A (2011) Dietary fiber intake and mortality in the NIH-AARP diet and health study. Arch Intern Med. 171: 1061-1068

Patron NJ, Smith AM, Fahy BF, Hylton CM, Naldrett MJ, Rossnagel BG, Denyer K (2002) The altered pattern of amylose accumulation in the endosperm of low-amylose barley cultivars is attributable to a single mutant allele of granule-bound starch synthase I with a deletion in the 5′-non-coding region. Plant Physiol. 130(1): 190-198

Prasopsunwattana N, Omary MB, Arndt EA, Cooke PH, Flores RA, Yokoyama W, Toma A, Chongcham S, Lee SP (2009) Particle size effects on the quality of flour tortillas enriched with whole grain waxy barley. Cereal Chem. 86: 439-451

PubMed Health (2011) U.S> National Library of Medicine. (http://www.ncbi.nlm.nih.gov/pubmedhealth/PMH0004546/) Verified February 13, 2013.

Quinde Z, Ullrich SE, Baik BK (2004) Genotypic variation in color and discoloration potential of barley-based food products. Cereal Chem. 81: 752-758

Quinde-Axtell Z, Ullrich SE, Baik BK (2005) Genotypic and environmental effects on color and discoloration potential of barley in food products. Cereal Chem. 82: 711-716

Rey JI, Hayes PM, Petrie SE, Corey A, Flowers M, Ohm JB, Ong C, Rhinhart K, Ross AS (2009) Production of dryland barley for human food: Quality and agronomic performance. Crop Sci. 49(1): 347-355

Rieder A, Holtekjølen AK, Sahlstrøm S, Moldestad A (2012) Effect of barley and oat flour types and sourdoughs on dough rheology and bread quality of composite wheat bread. J Cereal Sci. 55(1): 44-52

Risgaard H (2012) Perler fra Skærtoft Mølle. Politikens Forlag, København, Danmark

Robertson JA, Majsak-Newman G, Ring SG, Selvendran RR (1997) Solubilisation of mixed linkage (1→ 3),(1→ 4) β-D-glucans from barley: effects of cooking and digestion. J Cereal Sci. 25(3): 275-283

Ross AS (2011) (http://blogs.oregonstate.edu/deliciousnessw09/2011/09/29/more-kneading-conference-west-photos/) Verified February 13, 2013.

Ross AS (2013) Whole-grain and fiber-enriched noodles. In: Delcour J.A. and Poutanen K. (eds.) Wholegrain and fiber enriched foods: improving quality. Woodhead Publishing. In Press.

Ross AS and Bettge AD (2009) Passing the test on wheat end-use quality. In: Carver B. (ed.) Wheat: science and trade. Wiley-Blackwell Publishing, pp 455-493

Salamini F, Özkan H, Brandolini A, Schäfer-Pregl R, Martin W (2002) Genetics and geography of wild cereal domestication in the Near East. Nat Rev Genet. 3(6): 429-441

Savin R, Stone PJ, Nicolas ME, Wardlaw IF (1997) Effects of heat stress and moderately high temperature on grain growth and malting quality of barley. Aust J Agr Res. 48: 615-624

Shimizu C, Kihara M, Aoe S, Araki S, Ito K, Hayashi K, Watari J, Sakata Y, Ikegami S (2008) Effect of high β-glucan barley on serum cholesterol concentrations and visceral fat area in Japanese men - A randomized, double-blinded, placebo-controlled trial. Plant Food Hum Nutr. 63: 21-25

Skendi A, Biliaderis CG, Papageorgiou M, Izydorczyk MS (2010) Effects of two barley β-glucan isolates on wheat flour dough and bread properties. Food Chem. 119: 1159-1167

Slavin JL (2005) Dietary fiber and body weight. Nutrition. 21: 411-418

Slavin J, Tucker M, Harriman C, Jonnalagadda SS (2013) Whole Grains: Definition, Dietary Recommendations, and Health Benefits. Cereal Food World. 58(4): 191-198

Slukova M, Velebna N, Krejcirova L, Honcu I, Budilova E (2012) The Advanced Approaches to Nutritional and Breadmaking Quality of Wheat, Barley and Rye Flour. Journal of Food Science and Engineering. 2(4): 218-226

Sullivan P, Arendt E, Gallagher E (2013) The increasing use of barley and barley by-products in the production of healthier baked goods. Trends Food Sci Tech. 29(2): 124-134 (http://www.sciencedirect.com/science/article/pii/S0924224412002130)

Svihus B and Gullord M (2002) Effect of chemical content and physical characteristics on nutritional value of wheat, barley and oats for poultry. Anim Feed Sci Tech. 102: 71-92

Szczodrak J, Czuchajowska Z, Pomeranz Y (1992) Characterization and estimation of barley polysaccharides by near-infrared spectroscopy. II: Estimation of total β-D-glucans. Cereal Chem. 69(4): 419-423

Szűcs P, Skinner JS, Karsai I, Cuesta-Marcos A, Haggard KG, Corey AE, Chen TH, Hayes PM (2007) Validation of the VRN-H2/VRN-H1 epistatic model in barley reveals that intron length variation in VRN-H1 may account for a continuum of vernalization sensitivity. Mol Genet Genomics. 277(3): 249-261

Taketa S, Amano S, Tsujino Y, Sato T, Saisho D, Kakeda K, Takeda K (2008) Barley grain with adhering hulls is controlled by an ERF family transcription factor gene regulating a lipid biosynthesis pathway. P Natl Acad Sci USA. 105(10): 4062-4067

Taketa S, Kikuchi S, Awayama T, Yamamoto S, Ichii M, Kawasaki S (2004) Monophyletic origin of naked barley inferred from molecular analyses of a marker closely linked to the naked caryopsis gene (nud). Theor Appl Genet. 108(7): 1236-1242

Thondre PS and Henry CJK (2009) High-molecular-weight barley β-glucan in chapatis (unleavened Indian flatbread) lowers glycemic index. Nutr Res. 29: 480-486

Tiwari U and Cummins E (2011) Meta-analysis of the effect of β-glucan intake on blood cholesterol and glucose levels. Nutrition. 27: 1008-1016

Tiwari U, Cummins E, Sullivan P, Flaherty J, Brunton N, Gallagher E (2011) Probabilistic methodology for assessing changes in the level and molecular weight of barley β-glucan during bread baking. Food Chem. 124(4): 1567-1576

Topping DL and Clifton PM (2001) Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. Physiol Rev. 81(3): 1031-1064

Tosh SM, Brummer Y, Miller SS, Regand A, Defelice C, Duss R, Wolever TMS, Wood PJ (2010) Processing affects the physicochemical properties of β-glucan in oat bran cereal. J Agr Food Chem. 58: 7723-7730

Turnbull KM and Rahman S (2002) Endosperm texture in wheat. J Cereal Sci. 36(3): 327-337

van Amelswoort JMM and Westrate JA (1992) Amylose amylopectin ratio in a meal affects postprandial variables in male volunteers. Am J Clin Nutr. 55: 712-718

Vavilov NI (1951) The origin, variation, immunity and breeding of cultivated plants. Soil Sci. 72(6): 482

Verardo V, Gomez-Caravaca AM, Messia MC, Marconi E, Caboni MF (2011) Development of functional spaghetti enriched in bioactive compounds using barley coarse fraction obtained by air classification. J Agr Food Chem. 59: 9127-9134

Vitaglione P, Lumaga RB, Montagnese C, Messia MC, Marconi E, Scalfi L (2010) Satiating effect of a barley β-glucan-enriched snack. J Am Coll Nutr. 29: 113-121

Western Regional Climate Center. SOD USA Climate Archive. Available at http://www.wrcc.dri.edu/summary/ (verified Apr. 23, 2014).

Wolever TMS, Tosh SM, Gibbs AL, Brand-Miller J, Duncan AM, Hart V, Lamarche B, Thomson BA, Duss R, Wood PJ (2010) Physicochemical properties of oat β-glucan influence its ability to reduce serum LDL cholesterol in humans: A randomized clinical trial. Am J Clin Nutr. 92: 723-732

Wolfe MS, Baresel JP, Desclaux D, Goldringer I, Hoad S, Kovacs G, Löschenberger F, Miedaner T, Østergård H, van Bueren ETL (2008) Developments in breeding cereals for organic agriculture. Euphytica. 163(3): 323-346

Wood PJ, Beer MU, Butler G (2000) Evaluation of role of concentration and molecular weight of oat β-glucan in determining effect of viscosity on plasma glucose and insulin following an oral glucose load. Brit J Nutr. 84(01): 19-23

Wood PJ, Weisz J, Beer MU, Newman CW, Newman RK (2003) Structure of (1→ 3)(1→ 4)-β-d-glucan in waxy and nonwaxy barley. Cereal Chem. 80(3): 329-332

Xue Q, Wang L, Newman RK, Newman CW, Graham H (1997) Influence of the hulless, waxy starch and short-awn genes on the composition of barleys. J Cereal Sci. 26(2): 251-257

Zhang G, Chen J, Wang J, Ding S (2001) Cultivar and environmental effects on (1→ 3, 1→ 4)-β-D-glucan and protein content in malting barley. J Cereal Sci. 34(3): 295-301

Zheng GH, Rossnagel BG, Tyler RT, Bhatty RS (2000) Distribution of β-glucan in the grain of hull-less barley. Cereal Chem. 77(2): 140–144

Zheng MY, Weng Y, Liu W, Konzak CF (2002) The effect of ovary-conditioned medium on microspore embryogenesis in common wheat (*Triticum aestivum* L.). Plant Cell Rep. 20: 802–807

Zhong S, Dekkers JC, Fernando RL, Jannink JL (2009) Factors affecting accuracy from genomic selection in populations derived from multiple inbred lines: a barley case study. Genetics. 182(1): 355-364

Zobel RW, Wright MJ, Gauch HG (1988) Statistical analysis of a yield trial. Agron J. 80: 388–393