Project Title: Accelerated development of two-row facultative/winter malting barley

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Executive Summary

In order to assist AMBA in meeting its mission and to realize its primary objectives, OSU develops facultative/winter doubled haploid 2-row malting barley varieties. These will provide the malting and brewing industries with an abundant supply of high quality malting barley meeting the quality specifications of AMBA members. These varieties will have yield potential, making them attractive alternatives to competing crops. Our work is conducted within a larger framework of developing doubled haploid molecular breeding tools that will benefit all barley breeders working to advance the AMBA causes of mitigating risks and increasing acceptance rates.

The major issue for the OSU program is ensuring rapid and efficient development of facultative/winter 2-row malting barley varieties. Our major objectives are to use doubled haploids, molecular breeding tools, and collaborative phenotyping to quickly and efficiently address this issue.

One-year objectives and outputs:

- Submit promising new varieties to the AMBA approval system: The two OSU AMBA Plant Scale entries (OR-W1 and OR-W2) have attractive, balanced malt profiles for adjunct brewing. Nitrogen management and changes in malting protocol could make them (and particularly OR-W2) of interest to all-malt brewers. Agronomic performance is very good, particularly under irrigated conditions. Fungicides will be required in the higher rainfall areas of the Pacific Northwest for both the checks and the Plant Scale candidates. Winter hardiness is generally superior to Charles and comparable to Endeavor. These selections, and others, are in currently in regional testing in OSU-coordinated trials. They are also included in the 2015-2016 Winter Malting Barley Trial, Winter Barley Germplasm Nursery, and Uniform Barley Winter Hardiness Nursery. Both OR-W1 and OR-W2 are winters – they require vernalization.
- Develop new germplasm: Generated ~1,000 new facultative/winter doubled haploids and advanced them to regional/advanced/ preliminary trials. The malting quality profiles of these selections are promising, with profiles that could be of interest to adjunct and all-malt brewers.
- Target facultative growth habit: Initiated systematic characterization of facultative growth habit in all breeding material in the greenhouse in 2015 and continued in 2016. Will test facultative types in fall and spring-sown plantings beginning in 2016.
- Develop germplasm meeting all-malt brewer specifications: Developed doubled haploids specifically to meet these quality specifications.
- Increase and sustain doubled haploid production: Established a DH production facility that is meeting the needs of our own program and those of other programs, on a cost-recovery basis. Beginning in fall, 2016 the lab will focus exclusively on our own crosses and crosses made within the framework of grant-funded collaborations.

Most significant accomplishments:

- Moving OR-W1 and/or OR-W2 towards release as varieties.
- New doubled haploids submitted for AMBA evaluation and assessment in regional nurseries.
- Transfer of disease resistance and the lodging resistant, semi-dwarf growth habit to facultative/winter 2-row malting barley.
- Systematic introgression of European winter 2-row malting barley alleles into U.S. germplasm.
- Generating interest in winter/facultative barley throughout the barley research and production communities.

Detailed Report on Objectives, Methodology and Results – AMBA Funded Project

Objectives and Expected Benefits:

Our objective is to develop superior varieties that meet AMBA specifications based on an understanding of the genetic basis of target traits. In winter/facultative barley, our primary traits of interest are: malting quality, productivity, winter hardiness and disease resistance. All our efforts are directed at facultative/winter 2-row. The expected benefit is assisting AMBA in meeting its mission and primary objectives.

Methodology:

- All germplasm we develop is doubled haploid.
- Corvallis, OR is our principal test site. As germplasm advances it is tested regionally, nationally, and internationally. The 2015/2016 nurseries are summarized in Table 1.
- Malting quality assessments are conducted by the USDA/ARS and Dr. Cynthia Henson and colleagues collaborate on additional quality assays. Rich Joy at Great Western Malting provides additional malting quality data on special projects.
- Progress in our program depends on extensive collaboration. Dr. Kevin Smith, University of Minnesota is a key cooperator. BARI has provided agronomic testing at Fort Collins, Colorado and test sites in South Idaho. Dr. Gonghse Hu (USDA/ARS; Aberdeen, Idaho) provides data from Aberdeen. Dr. Juliet Marshall includes our advanced lines in the Idaho Extension nurseries. Dr. Kevin Murphy at Washington State University grows our material at Pullman, Washington. At Pendleton, Oregon we work with Dr. Stephen Machado and at Hermiston, Oregon with Matt Kolding. Dr. David Hole, Utah State, screens varying numbers of lines at Logan, Utah. We exchange germplasm with European companies.

Results:

Complete agronomic and quality data for OSU trials/germplasm are online at (http://barleyworld.org/breeding-genetics/data). In the interest of space, in this report we provide only summary data on our AMBA Plant Scale candidates and on the most promising new doubled haploids in Qualifying/Preliminary review. OR-W1 and OR-W2 are compared to the checks (Charles and Endeavor) for malting quality traits (Table 2) and agronomic traits (Tables 3a- 3c). Wintmalt is not included in these summaries as most data is from trials which precede its inclusion.

In terms of malting quality, both OR-W1 and OR-W2 have a higher average percentage of plump seed than the checks and a better range of values than Endeavor, which had

exceptionally thin seed at some locations. The average grain proteins for OR-W1 and OR-W2 are slightly lower than the checks, with similar ranges. The averages and ranges for malt extract are better for both OR-W1 and OR-W2 than either of the checks. OR-W1 had the highest wort protein and S/T; OR-W2 was the lowest. In terms of diastatic power, OR-W1 had an average value comparable to the checks, with a slightly wider range of values, whereas OR-W2 was lower on average and in the range of values. There was a similar pattern for alpha amylase. The average wort beta glucans for both selections were substantially lower than for the checks, and the ranges were closer to AMBA specifications than for the checks. OR-W1 had the highest FAN, in terms of both average and range. Considering all available data, the two OSU experimental lines have higher malt extracts, lower wort beta glucans, similar grain protein levels and enzymes, and substantially higher percentages of plump seed than the checks.

The yield data (Table 3a) are broken out by three categories for the Pacific Northwest: high rainfall, irrigated, and dryland. There was one environment of data from the Midwest (St. Paul, Minnesota). On average, OR-W1 was the highest yielding of the four, followed by Endeavor, OR-W2, and Charles. The OSU experimental lines and checks were most productive under irrigated conditions in south Idaho, with OR-W1 yielding over 7,500 lbs/acre. Under irrigation, OR-W2 was similar to Endeavor and 500 lbs/acre better than Charles. Under dryland conditions, Charles had the highest yield. Under high rainfall conditions, the two OSU selections had higher yields than both Endeavor and Charles. However, these yields are depressed due to the effects of diseases (Table 3c). The use of fungicides is needed in these environments. Endeavor had the highest average test weight, followed closely by OR-W1 and OR-W2 (Table 3b). Charles was a distant fourth. For kernel plumpness, however, OR-W2 and OR-W1 were the highest, followed by Charles and Endeavor. Under irrigated and dryland conditions, only Endeavor did not exceed 90% plump. Under high rainfall all four failed to meet the specification. The expectation is that with fungicide applications, yield, test weight, and plumpness will all increase under high rainfall conditions. In most trials contributing to this summary, there was 100% survival. However, where there was differential winter injury, OR-W2 had the highest survival, followed by OR-W1, Endeavor, and Charles. Heading dates are very similar for all varieties. Endeavor was the tallest variety, with the others being fairly similar in plant height. Lodging was a minor issue with Endeavor being most susceptible and OR-W1 the least. Brackling is a visual estimate of the percentage of straw breakage, on a plot basis. Both experimental selections were superior to the checks for this trait. All four entries are resistant/tolerant of stripe rust and susceptible to scald and leaf rust. The latter was formerly a minor pathogen in the Pacific Northwest – it is now a threat. These diseases are most common under high rainfall conditions, and as previously stated, fungicide protection of the checks, OR-W1, and OR-W2 will be required when these diseases are present. Considering all available data, the two experimental selections are generally equal to, or superior to the checks, for most agronomic traits.

Data on Qualifying/Preliminary doubled haploid selections are presented in Tables 4a-4b. There are higher levels of disease resistance, superior malting quality profiles, and many of the new entries are semi-dwarf types. This shorter, stiffer straw will maximize yield potential under irrigated and high rainfall conditions.

Other Barley Research and Future Direction of Program

In addition to winter malting barley development, the Oregon Barley Project is engaged in a number of other endeavors:

- Applying the results of winter hardiness (GWAS) conducted under the auspices of the TCAP.
- Genomic selection for winter hardiness and malting quality.
- Testing the hypothesis that barley can contribute to beer flavor.
- Implementing a malt lab whose primary function will be production and analysis of ~ 200 lb. batches of malt from advanced lines and new varieties.
- Winter barley for human nutrition.
- Genetic dissection of quantitative resistance to barley stripe rust.
- Breeding for UG99 stem rust resistance.
- Breeding for hooded forage barley varieties.

In the future, the Oregon Barley Program will continue its dual roles of stimulating economic development and contributing to the body of fundamental knowledge.

Project Personnel

- Patrick Hayes, Professor
- Tanya Filichkin, Senior Research Assistant
- Scott Fisk, Research Assistant
- Laura Helgerson, Research Assistant
- Dustin Herb, Graduate Research Assistant (PhD). Thesis research focuses on GWAS for low temperature tolerance-related traits and barley contributions to beer flavor.
- Javier Hernandez, Graduate Research Assistant (PhD). Thesis research focuses on GWAS, genomic prediction and genomic selection for facultative malting barley.

- Araby Belcher graduated in 2015. Her TCAP-supported research involved GWAS for malting quality, agronomic traits, and disease resistance. Manuscripts are in preparation.

Publications (2015-2016)

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Table 1. Oregon State University barley nurseries: 2015-2016. The summary is divided into “Malting”, “Food”, “Forage/Feed”, and “Genetics”. In reality, these areas of endeavor are tightly integrated. This is especially true for feed barley as we generally don’t breed specifically for this end-use however feed barley lines may stem from any of the other breeding areas. The emphasis of the program is on facultative growth habit in order to provide maximum flexibility in planting date (for growers) and breeding (for research and variety development).

Malting Barley

Overview:
Number of advanced and fixed lines: 679

- 217 in yield trial plots
- 462 in single rows or mini-plots (doubled haploids)

Details:

Oregon Winter/Facultative Barley Elite Yield Trial

• Corvallis, OR	Fall-planted	30 entries, 3 rep, RCBD
• Mt. Vernon, WA	Fall-planted	30 entries, 3 rep, RCBD
• Logan, UT	Fall-planted	30 entries, 3 rep, Lattice
• Aberdeen, ID	Fall-planted	30 entries, 3 rep
• Wooster, OH	Fall-planted	30 entries, 3 rep
• Rupert, ID	Fall-planted	30 entries, 2 rep
• St. Paul, MN	Fall-planted	30 entries, 2 rep
• Ithaca, NY	Fall-planted	30 entries, 2 rep
• Lebanon, OR	Fall-planted	30 entries, unreplicated

Oregon Malting Barley Advanced Line Yield Trial

• Corvallis, OR	Fall-planted	47 entries, 2 rep, RCBD
• Logan, UT	Fall-planted	44 entries, 3 rep, Lattice
• Lebanon, OR	Fall-planted	44 entries, unreplicated
• St. Paul, MN	Fall-planted	44 entries, observation rows

Oregon Malting Barley Preliminary Yield Trial

• Corvallis, OR	Fall-planted	63 entries, unreplicated
• St. Paul, MN	Fall-planted	63 entries, observation rows

Oregon Facultative Barley Yield Trial

• Madras, OR*	Spring-planted	33 entries, 2 rep, RCBD

Oregon Promise Yield Trial

• Lebanon, OR	Winter-planted	26 entries, 2 rep, RCBD
• Madras, OR*	Spring-planted	26 entries, 2 rep, RCBD

Winter Malting Barley Trial (US and International Cooperators)

• Corvallis, OR	Fall-planted	28 entries, 3 rep, RCBD

Winter Barley Germplasm Nursery (US Cooperators)

• Corvallis, OR	Fall-planted	20 entries, 2 rep, RCBD

AMBA Drill Strips

• Corvallis, OR	Fall-planted	29 entries (OSU & USDA-ARS)
• Lebanon, OR	Fall-planted	29 entries (OSU & USDA-ARS)
• Pendleton, OR	Fall-planted	12 entries (OSU & USDA-ARS)
• Pullman, WA	Fall-planted	8 entries
• Aberdeen, ID	Fall-planted	6 entries
• Rupert, ID	Fall-planted	3 entries

Observational Drill Strips/Seed Increase

- Hermiston, OR — Fall-planted — 2 entries

Oregon Promise Drill Strips

- Madras, OR* — Spring-planted — 4 entries

Purification Head Rows

- Corvallis, OR — Fall-planted — 2 entries, 432 rows each

Malt Doubled Haploid Mini-Plots

- Corvallis, OR — Fall-planted — 312 lines, unreplicated w/checks

Malt Doubled Haploid Single Rows

- Corvallis, OR — Fall-planted — 150 lines, unreplicated w/checks

2-row Spring Barley Fertility Trial (BOBB)

- Willamette Valley — Winter-planted — 3 entries, 3 treatments, 4 rep, RCBD
- Klamath Basin* — Spring-planted — 3 entries, 3 treatments, 4 rep, RCBD
- Grande Ronde Valley* — Spring-planted — 3 entries, 3 treatments, 4 rep, RCBD

*Not yet planted as of 2/29/16

Food Barley

Overview:

Number of advanced and fixed lines: 61

- 55 in yield trial plots
- 6 mini-plots (doubled haploids)

Details:

Oregon Food Barley Elite Yield Trial

- Corvallis, OR — Fall-planted — 23 entries, 3 rep, RCBD
- Lebanon, OR — Fall-planted — 23 entries, Unreplicated
- Mt. Vernon, WA — Fall-planted — 23 entries, 3 rep, RCBD
- St. Paul, MN — Fall-planted — 23 entries, 2 rep
- Ithaca, NY — Fall-planted — 23 entries, 2 rep

Oregon Food Barley Preliminary Yield Trial

- Corvallis, OR — Fall-planted — 18 entries, unreplicated

Black Beaut Yield Trial

- Madras, OR* — Spring-planted — 14 entries, 2 rep, RCBD

Observational Drill Strips/Seed Increase

- Corvallis, OR — Fall-planted — 2 entry
- Lebanon, OR — Fall-planted — 2 entry
- Pendleton, OR — Fall-planted — 1 entry
- Pullman, WA — Fall-planted — 1 entry

Food Mini-Plots

- Corvallis, OR — Fall-planted — 6 lines, unreplicated

*Not yet planted as of 2/22/16

Forage & Feed^

Overview:

Number of advanced and fixed lines: 54

- 44 in yield trial plots/drill strips

- 10 in mini-plots (doubled haploids)

Details:
On-Farm Strip Trial (Morrow County)

• Jepsen Farm	Fall-planted	8 entries, 4 rep, RCBD
• Starvation Farm	Fall-planted	7 entries, 4 rep, RCBD

Hooded Barley Yield Trial

• Corvallis, OR	Fall-planted	32 entries, 2 rep, RCBD

Observational Drill Strips/Seed Increase

• Corvallis, OR	Fall-planted	4 entries
• Pendleton, OR	Fall-planted	3 entries
• Hermiston, OR	Fall-planted	3 entries
• Pullman, WA	Fall-planted	3 entries

Purification Head Rows

• Corvallis, OR	Winter-planted	1 entries, 108 rows

Hooded Mini-Plots

• Corvallis, OR	Fall-planted	10 lines, unreplicated

^Not an inclusive list as feed lines are integrated within other breeding classes. All lines grown have potential as a possible feed variety.

Genetics

Overview:
Number of advanced and fixed lines: 579

- 579 in single, double or mini-plots

Details:
Barley Stripe Rust Nursery (Corvallis); Winter-planted

• Barley Stripe Rust Screening Trial	73 entries, 2 reps, RCBD
• FacWin 2-row TP	139 entries, 2 reps, RCBD
• UG99 Germplasm Array	94 entries, 2 reps, RCBD
• 95SR316A/GZ Population	165 entries, 2 reps, RCBD
• Uniform Barley Winter-hardiness Nursery	17 entries, 2 reps, RCBD
• BARI BSR Nursery	50 entries, 2 reps, RCBD

Low Temperature Tolerance Mini-Plots

• Corvallis, OR	Fall-planted	24 entries, unreplicated

UG99 Mini-Plots

Corvallis, OR	Winter-planted	17 entries, unreplicated

Other

Adaptive Symbiotic Technologies Drill Strips

• Corvallis, OR	Fall-planted	3 entries, 2 Trtmnts, 8 rep, Split Plot
• Lebanon, OR	Fall-planted	3 entries, 2 Trtmnts, 8 rep, Split Plot

Table 2. Malting quality of OSU AMBA Plant Scale candidates vs. checks across years and locations. *Ranges are in italics*. Wintmalt is not included in the Table 3a – 3c summaries because it was not present in all trials. It will be included in all future trials.

Variety/ Selection	Plump seed (% on 6/64)	Barley protein (%)	Malt extract (%)	Wort protein (%)	S/T (%)	Diastatic power (^{0}ASBC)	Alpha-amylase (20^0 DU)	Beta glucan (ppm)	FAN (ppm)
OR-W1	94 *79-99*	11.3 *10-14*	82.2 *80-85*	5.7 *5-7*	55 *47-66*	160 *92-216*	102 *49-126*	66 *21-193*	287 *250-394*
OR-W2	95 *85-98*	11.5 *11-14*	82.1 *81-84*	5.1 *4-5*	49 *41-60*	138 *92-170*	80 *45-105*	80 *43-124*	245 *195-299*
Charles	88 *79-99*	12.0 *11-15*	80.6 *77-83*	5.4 *5-6*	49 *43-56*	149 *112-198*	89 *61-111*	194 *59-291*	246 *186-310*
Endeavor	81 *57-96*	11.7 *10-14*	81.1 *80-83*	5.3 *5-6*	51 *44-58*	160 *108-193*	93 *51-112*	201 *51-422*	239 *196-292*
Station years	11	11	12	12	12	12	12	12	12

Table 3a. Yield of OSU AMBA Plant Scale candidates vs. checks across years and locations.

Variety/ Selection	Yield High rainfall (lbs/acre)	Yield Dryland (lbs/acre)	Yield Irrigated (lbs/acre)	Yield Midwest (lbs/acre)	Yield Grand Mean (lbs/acre)
OR-W1	5189	4890	7524	4205	5452
OR-W2	4892	4492	7133	3954	5118
Charles	4127	4946	6630	4050	4938
Endeavor	4785	4762	7154	4309	5253
Station years	9	3	8	1	21

Table 3b. Test weight and % plump seed of OSU AMBA Plant Scale candidates vs. checks across years and locations. *Note: The % plump data in this table are for all environments; those reported in Table 2 are only for samples submitted for malt analysis.*

Variety Selection	Test weight High rainfall (lbs/bu)	Test weight Dryland (lbs/bu)	Test weight Irrigated (lbs/bu)	Test weight Grand Mean (lbs/bu)	% Plump High rainfall (6/64)	% Plump Dryland (6/64)	% Plump Irrigated (6/64)	% Plump Grand Mean (6/64)
OR-W1	49	52	51	51	81	94	95	90
OR-W2	48	50	51	50	86	90	95	90
Charles	44	50	49	48	77	92	92	87
Endeavor	49	53	53	52	67	79	89	78
Station years	8	3	6	17	8	3	6	17

Table 3c. Winter survival (where there was differential winter injury), Heading date, Plant height, Lodging, Brackling, Scald, Leaf rust, and Stripe rust data for OSU AMBA Plant Scale candidates vs. checks across years and locations. **Only noted in high rainfall environments.*

Variety/ Selection	Winter survival (%)	Heading date (DOY)	Plant height (in)	Lodging (0-9)	Brackling* (%)	Scald* (%)	Leaf rust* (%)	Stripe Rust* (%)
OR-W1	80	136	35	1	29	51	85	7
OR-W2	92	136	33	2	27	63	78	0
Charles	79	135	36	2	56	78	77	0
Endeavor	79	134	39	3	39	45	77	6
Station years	12	11	20	11	6	5	3	5

Table 4a. Malting quality of OSU Qualifying/Preliminary selections vs. checks across years and locations.

Variety/ Selection	Kernel Plumpness (on 6/64")	Grain Protein (%)	Malt Extract (%)	Wort Protein (%)	S/T (%)	Diastatic Power (°L)	Alpha Amylase (20° DU)	Beta-Glucan (ppm)	FAN (ppm)
DH130718	98.5	12.9	80.0	5.34	43.8	167	82.0	77	255.1
DH120412	96.0	12.1	79.8	5.27	45.9	217	73.1	55	260.4
DH130004	91.1	12.7	79.3	5.78	47.4	180	74.8	51	300.3
DH 10.1044	92.1	11.1	80.2	3.89	37.3	205	42.7	93	148.2
DH120228	94.1	11.3	81.9	5.29	49.8	186	111.3	39	248.7
DH120276	94.1	11.8	80.3	5.08	46.0	170	94.4	67	231.8
DH130939	95.4	12.1	80.6	5.21	45.7	165	101.9	141	258.2
DH130765	98.1	14.0	80.2	6.15	45.7	241	83.5	53	286.5
DH130910	97.0	12.0	80.0	4.55	39.6	177	39.4	81	183.5
10.0925	93.5	10.5	79.8	3.60	37.1	91	42.3	108	143.0
Charles	91.5	11.2	79.9	5.09	48.2	148	93.7	178	233.7
Endeavor	73.2	10.7	80.6	5.06	50.9	170	99.1	177	234.3
Wintmalt	93.3	11.3	79.2	4.04	37.4	155	53.7	77	169.7
Station Years	3	3	3	3	3	3	3	3	3

Table 4b. Agronomic data of OSU Qualifying/Preliminary selections vs. checks across years and locations.

Variety/ Selection	Yield bu/acre	Lodging (1-9†)	Brackling (%)	Plant Height Inches	Scald (%)	Stripe Rust (%)	Survival (%)
DH130718	149	1	6	39	2	0	68
DH120412	131	1	20	41	4	13	80
DH130004	107	1	8	33	8	28	82
DH 10.1044	127	1	19	38	41	0	47
DH120228	132	5	35	43	9	2	58
DH120276	107	1	21	43	18	10	53
DH130939	126	1	22	41	11	7	62
DH130765	121	1	8	37	3	0	57
DH130910	109	2	13	45	9	7	94
10.0925	112	1	22	37	27	2	89
Charles	92	3	62	40	86	0	60
Endeavor	93	3	31	40	74	4	52
Wintmalt	94	2	24	39	47	11	76
Station Years	4	4	3	4	3	3	3