

American Malting Barley Association, Inc.
2015 Research Grant Program
Executive Summaries
(July 1, 2015 – June 30, 2016)
Total Funding = \$576,610

EASTERN VARIETY DEVELOPMENT & RESEARCH

Northeast Malting Barley Production

Heather Darby, University of Vermont

The localvore movement has created and expanded markets for farmers in the Northeast, to the point where demand for local grains exceeds supply. More recently, public interest in sourcing local foods has extended into beverages, and the current demand for local brewing and distilling ingredients is quickly increasing. One new market that has generated interest of both farmers and end-users is malted barley. This only stands to reason since the Northeast alone is home to over 175 microbreweries and 35 craft distillers. Additionally, in New York a provision in the legislation calls for 90% of the hops and 90% of the ingredients in local beer to be New York grown by 2024. The same provision has just been proposed in the state of VT. These activities have obviously significantly increased the demand for local grains and malt. Until recently local malt was not readily available to brewers or distillers. However a rapid expansion of the fledgling malting industry will hopefully give farmers new markets and end-users hope of readily available malt. Six new malt houses in VT and NY have opened their doors since 2014. In addition, established malt houses such as Valley Malt (Hadley, MA) and Maltarie Frontenac (Thetford Mines, Qc) have expanded their production capacities to meet the demand from brewers and distillers. In addition, larger malting companies such as Canada Malting Company have also expressed interest in further expanding their local product development, which would create even more space for additional farms to enter the market. To date, the operating maltsters struggle to source enough local grain to match demand for their product. Those businesses yet to open have expressed concern that local grain quantities will not be available to support their new operations. In addition to short supplies, the local malt barley that is available often does not meet the rigid quality standards for malting.

Farmers in the Northeast have long grown barley for animal feed on dairy farms, but they lack local knowledge, research information, and resources on how to produce barley that meet malting specifications. Small grains are held to very rigorous standards in the malting industry. With any new crop there is a steep learning curve and therefore farmers must learn how to implement best agronomic practices combined with proper harvest, drying, and storage techniques to meet these malt standards. Therefore the goal of this project is to enhance the capacity of farmers to produce high quality malt barley to meet the increasing demand from local brewers and distillers, and ultimately consumers.

There are many barriers to producing high quality grain in the Northeast. However preliminary data developed by the project team illustrates that malting quality barley can be produced in the region (Darby, 2010; 2011; 2012). However there are still many challenges to overcome such as major crop losses to head scab as experienced in 2013 and 2014. Regional research is needed to develop optimal agronomic practices that will help farmers meet the exacting standards of malting quality small grains. Locally adapted variety, planting date, harvested, fertility and weed control recommendations would assist producers in increasing their marketable yields.

The first step is to develop variety trials that evaluate potential germplasm for the area. Variety trials will identify commercially available and heirloom varieties as well as experimental lines that might be suitable for malting and adapted to the Northeast. Successful crop production starts with choosing varieties that are well adapted to the specific growing conditions. Winter and spring malt barley trials conducted in VT illustrate that high yield and quality barley can be produced in the region. The trials also clearly indicate that few winter varieties are adapted to the region and that scab tolerant varieties will be extremely important for success of this crop. Further testing over multiple years and location is needed to provide adequate recommendations to farmers.

Development of Two and Six-rowed Winter Malt Barley for Virginia and the Southeastern US *Carl Griffey, Virginia Tech*

The overall objective of this project is to develop two and six-rowed winter malt barley varieties with acceptable malt quality, improved disease resistance and high yield potential to provide a uniform supply of winter malting barley for small and large scale malting and brewing industries. Success and expansion of the current winter malt barley industry in Virginia and the eastern U.S. will largely depend on efforts made in breeding, evaluation, and production of winter barley and also sound management practices and control of economically devastating diseases. The proposed research is designed to develop winter malt barley varieties that are adapted to the diverse regions in the eastern U.S. that have high quality, improved disease resistance, and high yield potential. To meet these objectives, we are conducting comprehensive breeding and genetics research supported by grant funds from Small Grain Boards (Virginia, Kentucky and Maryland) and the United States Wheat and Barley Scab Initiative (USWBSI). This AMBA project supports breeding activities such as making crosses, population development, trait evaluation, and breeding line selection directed towards the development of new winter malt barley varieties. This research will help initiate and subsequently expand winter malt barley production into areas where previous production has been considered too risky or not feasible. In addition, our research objectives all directly address the primary aim of the American Malting Barley Association (AMBA) by the development of improved winter malt barley varieties and providing adequate supply of high quality winter malting barley for Craft Breweries in Virginia and indeed the south eastern U.S.

Development of Winter Malting Barley for North Carolina and Adjoining States *David Marshall, USDA-ARS, Raleigh, NC*

In response to local and organic food production thrusts in North Carolina, D. Marshall began a winter, 2-row, malting barley variety development program in 2010. The goal of the proposed research is to assist in accelerating the development of winter malting barley for North Carolina and adjoining states. Our selection is unique in North Carolina because we select under both conventional and organic production practices. The organic selection is conducted at the Center for Environmental Farming Systems (CEFS) in Goldsboro, NC. A potential obstacle in the episodic nature of Fusarium Head Blight (FHB) impacting germplasm and varietal development. So, we began to screen all our germplasm in the USDA-ARS FHB mist nursery in Raleigh, NC. In the 2014-15 season, we initiated a nitrogen and growth regulator trial in Mills River, NC. We would like to continue the management trials in 2015-16.

MIDWEST VARIETY DEVELOPMENT & RESEARCH

Barley Improvement

Kevin Smith, University of Minnesota

The overall objective of this research is to develop new barley varieties with acceptable malt quality, improved disease resistance, and high yield potential. At present, we are developing spring and winter (facultative) six-row and two-row malting barley for the Midwest region. Our overall goal is to produce an array of barley varieties that provide maximum flexibility to growers, are adapted to Midwest fall and spring seeded cropping systems, and produce high quality malt. We make our advanced lines available to all interested researchers for testing in their regions. This research will directly assist in AMBA's mission to provide an adequate supply of high quality malting barley. This research is a cooperative effort of the Departments of Agronomy and Plant Genetics, Plant Pathology, and Research and Outreach Centers of the University of Minnesota. Specific breeding goals include high yield, enhanced lodging resistance, resistance to Fusarium head blight (FHB), net blotch, bacterial leaf streak (BLS), spot blotch, stem rust, and Septoria speckled leaf blotch (SSLB), and favorable malting and brewing characteristics. In addition, in our winter barley program, we seek to enhance winter hardiness and combine it with high yield and good malting quality. To meet these objectives, we are conducting a comprehensive breeding and genetics research effort funded by Minnesota Agricultural Experiment Station, state, and federal grants. This AMBA proposal is designed to support research activities not funded through other sources, to maximize the effectiveness of resources, and to insure that germplasm, information, and technological discoveries are translated rapidly toward the development of new malting barley varieties.

Management and Epidemiology of Barley Diseases

Ruth Dill-Macky, University of Minnesota

This is an applied research program directed at the management of the diseases of economic importance to the barley industry in Minnesota and the Upper Midwest. The project is instrumental in screening barley germplasm (6-row spring, 2-row spring and 2-row winter types) for resistance to diseases with economic impact, conducting annual surveys of commercial barley crops to monitor diseases, and conducting research to develop and deploy effective disease control practices. The screening of germplasm, conducted in collaboration with the Minnesota barley breeding program, serves both to identify progeny suitable for advancement and/or release as commercial malting barley cultivars and provides information for the genetic characterization of resistance and the development of markers that may be utilized in marker assisted selection (MAS). The diseases we are currently working with include; Fusarium head blight, net blotch, bacterial leaf streak, powdery mildew and barley yellow dwarf.

Monitoring barley crops in the Upper Midwest annually for the prevalence of diseases serves to identifying potential new and/or emerging threats to the industry and insure against the unexpected upsurge of diseases that may impact barley yield and/or quality. We anticipate that in this project we will conduct summer survey trips examining commercial barley crops for disease and making collections of representative pathogen isolates in the field. Our collection of barley pathogens is then utilized in resistance screening and in studies that examine the structure and diversity of pathogen populations.

In addition to providing services to the breeding program, this program conducts independent research aimed at the development of effective and efficient techniques for the identification of resistance to barley diseases. Studying the epidemiology of diseases and testing a variety of management options facilitates the implementation of effective disease control practices for the diseases of economic importance to barley. Thus we conduct field and greenhouse experiments to develop and evaluate chemical, biological and/or cultural control options for the management of barley diseases.

The studies undertaken in this project, and the development of new barley cultivars, are ongoing activities, the scope of which extend beyond a one-year project. Annual outcomes from this project can however be measured through the release of new malting quality barley varieties, through the numbers of lines screened for their reaction to various diseases, and through research outcomes published in peer-reviewed scientific journals and extension publications including the variety trails bulletin.

Breeding and Genetics of Six- and Two-rowed Malting Barley

Richard Horsley, North Dakota State University

The basic objective of the malting barley breeding program at North Dakota State University (NDSU) is to develop and release improved varieties acceptable to barley producers in the northern Great Plains and to those who use or process this barley. New crosses are made each year and progeny are constantly moved through the “breeding pipeline.” Obstacles that may impact our research include natural disasters such as extreme weather (e.g. drought, excess precipitation, excessive winds, etc.) that may directly affect research sites and our ability to obtain reliable data and/or harvest materials for advancement in the program. In each stage of the breeding process, progeny having desirable traits are identified and advanced for further testing. Because growers in the region have choices on which crops to grow on their farms, it is our responsibility to develop improved malting barley varieties that are competitive with other crops so barley is an economically viable choice for them to grow. Plant characters that receive the most attention are those that affect quality of the grain, resulting in losses to producers and end users. These characters include yield, grain protein, kernel plumpness, resistance to Fusarium head blight (FHB) and foliar diseases, and resistance to preharvest sprouting (PHS). Our barley-breeding project also conducts research that leads to improved farming systems that promote production of barley with acceptable malting quality.

Since the two- and six-rowed barley programs were consolidated in 2006, about 60% of the program’s efforts were dedicated towards development of six-rowed barley. Based on increased demand for two-rowed malting barley adapted for production in North Dakota, our program made more two-rowed crosses than six-rowed crosses in 2014. It is expected that a minimum of 60% of the entries in replicated yield trials in 2017 will be two-rowed. Our emphasis in both programs will continue to be developing new malting barley varieties for adjunct and all malt brewers with improved resistances to foliar diseases, Fusarium head blight, deoxynivalenol (DON) accumulation, and pre-harvest sprouting.

Genomic selection will be used to identify two-rowed F₅ lines in our Yuma, AZ winter-season nursery that are likely to have improved resistances to spot blotch, net-form net blotch, spot-form net blotch, and reduced DON accumulation; and reduced wort β -glucan concentration. Approximately 2,400 two-rowed lines will be genotyped in Dr. Bob Brueggeman’s using 384 markers. Identified lines with acceptable straw strength in the winter nursery will be advanced to preliminary yield trials at two locations in North Dakota in 2016.

Managing Diseases that Impact Malting Barley

Robert Brueggeman, North Dakota State University

Diseases are the most important factor limiting yield and quality of malting barley production in the upper Midwestern United States. To help ensure that diseases have less impact on an adequate supply of quality malting barley produced in the U.S. our research is focused on economically feasible methods of disease management. Thus, the primary goal of the Barley Pathology Program at

North Dakota State University (NDSU) is to conduct applied and basic research focused on the development of effective genetic resistance to important barley diseases. We are achieving this goal through collaborations with the NDSU, University of Minnesota, MillerCoors, USDA-Aberdeen, ID, USDA-Fargo, ND, Washington State University, and the University of Idaho barley-breeding program and research programs. We screen experimental germplasm against a range of SNP markers associated with economically important diseases and focus our genotyping collaborations on research that will ultimately result in better disease management. The NDSU Barley Pathology Program phenotypically screens material for resistance to fusarium head blight (FHB), spot form net blotch (SFNB), net form net blotch (NFNB), spot blotch (SB), stem rust (SR), leaf rust (LR) and bacterial leaf streak (BLS) in the greenhouse and field.

Our efforts also involve screening germplasm collections for new sources of resistance, conducting association mapping studies with these collections and developing targeted bi-parental mapping populations to characterize resistance, develop molecular markers and barley populations that will expedite the deployment of resistance. We are utilizing the genetic knowledge gained from these projects to isolate and characterize NFNB, SFNB, SB, and SR resistance genes in barley in order to elucidate the underlying molecular mechanisms determining resistance/susceptibility. The information gained from the functional analyses will allow for more intelligent deployment of resistance and has already provided “perfect markers” for marker assisted selection of Ug99 resistant barley lines.

Collections of pathogen populations are also being made to monitor virulence spectra as well as map and identify virulence genes to ensure that changes in the pathogen populations, that could threaten the effectiveness of deployed resistance, can be detected and addressed in a timely manner. In addition we are developing novel techniques utilizing next-generation sequencing technology to reduce the resource drain encountered when genotyping barley and pathogen populations. These tools have enhanced our ability to conduct research to characterize barley disease resistance genes and the corresponding virulence factors in the pathogens with the limited funding available.

Stem Rust Resistance Enhancement

Maricelis Acevedo, North Dakota State University

Wheat stem rust is an economically important disease of barley everywhere barley is grown. To date only two highly effective *P. graminis f. sp. tritici* (*Rpg*) resistance genes have been identified. Of those, only one, the *rpg4/Rpg5* locus provides resistance to the highly virulent stem rust races of the TTKS (Ug99) lineage (Steffenson et al 2009). Despite these resistances being effective against current stem rust pathogen races, it is alarming to think that the entire barley crop depends on two major resistances against a pathogen that is highly variable and constantly evolving. Newer races of the pathogen such as QCCJ in the 90's in the US and the emerging Ug99 lineage in Eastern Africa serve as a reminder of the vulnerability of the crop. Recently, evaluation of a double haploid population of a cross between the barley lines Q21861 (stem rust resistant) and SM89010 (stem rust seedling susceptible) identified a “hot-spot” for genes associated with an enhancement of stem rust resistance in the barley chromosome 2H (Moscou et al., 2011). An earlier study, utilizing a different mapping population had also identified a region associated with stem rust resistance in a second location of this chromosome (Druka et al., 2008). Surprisingly this genetic region seems to not been actively utilized in breeding programs in the past.

The objective of the proposed project, which started in 2012, is to incorporate these two genetic regions to expand the genetic basis of the stem rust resistance response in adapted barley cultivars with superior agronomic, malting, and brewing characteristics which already combine the *Rpg1* and

the *rpg4/Rpg5* resistances. By incorporation of this “resistance enhancer” we will be able to increase the resistance to Ug99 as well as local races. To be able to efficiently select and pyramid these resistances in breeding programs, highly reproducible genetic markers tightly associated with the new resistance enhancer regions need to be identified. This step is extremely important for an efficient incorporation of these genetic regions since their effect seems to only be noticeable in the presence of other genes with a larger effect on resistance. This project fits with AMBA’s mission and primary goals of developing improved varieties with enhanced, longer lasting and broader disease resistance, specifically Ug99 resistance.

Supporting Malting Barley Production Growth in Northern Michigan

Christian Kapp, Michigan State University

Craft brewing is one of the fastest growing industries in Michigan, with both an increase in production volume and establishment of new breweries. Michigan is considered by the Brewers Association to be a high growth state with an increase of 20% per year in the form of new breweries. Knowledge of how well current malting barley varieties perform within Michigan’s geographic region is currently being sought by both entrepreneurs forming malt house businesses, and growers who are interested in meeting malt house needs. Michigan’s climate is inherently challenging in terms of malting barley production with respect to rainfall during the ripening period and favorable conditions for plant disease.

This research proposal directly addresses AMBA’s primary mission of encouraging and supporting an adequate supply of high quality malting barley and also by increasing the understanding of malting barley. Outcomes from this proposal will lead to increased information on agronomic performance and quality characteristics of certain malt barley lines grown under Michigan’s climatic conditions. Current research has indicated that certain barley varieties perform better with respect to pre-harvest sprout. Further experimentation needs to be continued in order to determine if this is indeed a trend. Specific outputs generated will be agronomic, disease and malt quality characteristics that will be specific to variety. Long term outcomes of this project will lead to an overall increase of acreage within the state, specifically within northern Michigan.

WESTERN VARIETY DEVELOPMENT & RESEARCH

Pyramiding Genes for Disease Resistance

Lynn Gallagher, University of California, Davis

The barley breeding program at UC Davis will develop two-rowed germplasm which combines multiple disease resistances and malting quality with good agronomic performance. We are pyramiding genes conferring resistance to several viruses into a malting barley background. Resistance to three virus groups (ie: BYDV-PAV, CYDV-RPV, and BYDV-RMV) are a priority. Work will be done on the following: 1) add the genes *Yd2* and *Yd3* which while separately conferring tolerance together confer resistance to BYDV-PAV, the most common virus in this three virus group 2.) continue field selection for resistance to cereal yellow dwarf virus (CYDV-RPV) until molecular markers become available years from now, 3) develop populations to study the resistance to the BYDV-RMV species. The RMV species from Montana does severe damage to plants. RMV is found across the malting barley grain belt. 4) continue to screen elite germplasm developed for multiple disease resistances by CIMMYT in Mexico. Malting barley varieties will become more broadly adapted in North American production areas because of the addition of genes conferring disease resistance. The developed germplasm will be distributed to both private and public institutions which express an interest in using germplasm for breeding purposes.

Spring and Winter Malting Barley Development

Gongshe Hu, USDA-ARS, Aberdeen, ID

Our project mission is to 1) develop superior widely adapted spring and winter two-rowed and six-rowed malt barley cultivars, and 2) develop improved spring and winter barley germplasm for use by public and private barley breeding programs. In addition to cultivar and germplasm development, our work also provides valuable information to industry, private and public breeders, and barley producers via multi-location testing of lines from other breeding programs. Therefore we assist in meeting the mission of AMBA by the development of improved cultivars and germplasm, and assisting other breeding programs in their effort to release improved cultivars.

We are specifically addressing the development of improved winter and spring malting cultivars for the Intermountain West (IMW) areas of Idaho, Montana, Oregon, and Washington with the goal of releasing lines which not only meet industry standards for malt quality, but also exceed existing cultivars in agronomic performance, benefiting both the malting industry and the barley producer. Included in these agronomic characteristics are improved yield, lodging resistance, and disease resistance for both spring and winter cultivars. In addition, improved winter hardiness is a major objective, especially for the two-rowed winter program. AMBA funding allows us to evaluate lines at multiple locations leading to the development of cultivars with wide environmental stability for use across large growing areas.

Barley Breeding for Montana – Ensuring a Stable Malt Supply

Jamie Sherman, Montana State University

Montanan growers plant more barley acres than any other state. Successful barley crops in Montana are critical to the security of malting barley for end-users. Growers and end-users recognize the critical nature of barley for Montana and the country, since much of what is grown is used by stakeholders in other states. In fact, grower and end-user support ensured the continuation of the Montana barley breeding program at Montana State University with the hire of a new breeder in Jan. of 2015. Threats to producing malt barley in Montana include: 1) The movement of corn and soybean into Montana because they provide a safer higher economic return to growers. 2) Abiotic stresses, which are becoming more frequent and less predictable with climate change, reduce yield or cause barley to be rejected for malting, reducing growers' income. 3) Biotic stress patterns are changing with the climate and in some cases are invading Montana.

Montana State University Barley Breeding Program's primary mission is to develop new lines that ensure an adequate supply of high quality malting barley for the malting, brewing, distilling, and food industries, providing growers and the state with important income. Our primary objective is to enhance barley competitiveness by increasing yield and yield stability and improving quality stability, reducing risk to growers so they will produce malting barley. Another objective of the program is to identify, develop and utilize new traits in released varieties that will provide an economic advantage to growers or end-users.

Accelerated Development of Two-row Facultative/Winter Malting Barley

Patrick Hayes, Oregon State University

The goals of this research are to develop doubled haploid facultative/winter 2-row malting varieties adapted to the Pacific Northwest and to assist breeders throughout the nation by providing useful germplasm. The benefit to the malting and brewing industries will be a continuous supply of high

quality malting barley and the benefit to growers will be varieties that are more economically competitive with corn and wheat. Facultative varieties, which are capable of maximum cold tolerance when fall-planted but do not require vernalization and can therefore be spring-planted – offer growers and processors a risk management tool to deal with climate volatility.

The primary objective is to ensure a steady stream of AMBA-approved varieties, and the corresponding output from realizing this objective is a steady stream of varieties. To accomplish this objective, and ensure this output, we will continue our doubled haploid-based breeding program. This program involves repeated cycles of crossing, doubled haploid production, extensive phenotyping, and genotyping. Genomic prediction can be integrated into this accelerated breeding scheme. New germplasm – e.g. from other programs and genetics projects - such as the TCAP LTT project - is infused during each crossing season.

Aphid Resistant Malting Barley Germplasm Enhancement & Evaluation

Dolores Mornhinweg, USDA-ARS, Stillwater, OK

Whenever insect pests impact grain yield of barley, *Hordeum vulgare* (L.), they affect malting quality. Historically there are three yield limiting aphid pests of barley in the US, Russian wheat aphid, *Diuraphis noxia* (Kurdjumov), RWA; greenbug, *Schizaphis graminum* (Rondani), GB; and bird cherry-oat aphid, *Rhopalosiphum padi* (L.), BCOA. A newly introduced aphid pest, *Sipha maydis*, (SA) has been reported in New Mexico and Colorado. If established this aphid has the potential to become a serious threat to US wheat and barley. RWA is a pest in the malting barley production areas of the western US while BCOA is common to all barley production areas. SA is likely to establish in areas similar to RWA but could be a threat to all barley production areas.

The major cause of yield loss with RWA feeding is head trapping which results in reduced fertility and a severe decrease in plumpness of surviving seed. Not only is less grain available for malt, but the quality of that grain is greatly decreased. When RWAs feed on susceptible plants, the new leaves do not unroll and aphids build up in high numbers inside the unrolled leaves where they are protected from contact insecticides as well as natural parasites and predators, wind and rain. In years of severe or early infestation, chemical control can only be accomplished with repeated applications of systemic insecticides. These chemicals are not only expensive to the grower; they could quite possibly end up in malt produced from treated fields. Greenhouse seedling screening of the entire NSGC of barley resulted in identification of 116 unadapted RWA-resistant germplasm lines.

BCOA, most often considered a crop pest because of its ability to vector yield devastating BYDV, has been reported to cause yield reduction of up to 50% from aphid feeding alone. BCOA can damage spring barley and winter barley both in the fall and spring. Yield reductions are attributed to a reduction in seed number and seed weight and have been associated with BCOA damaged plants' inability to withstand environmental stress. BCOA feeding negatively impacts the root system as well as above ground growth and reduces winter hardiness of cereals. With new emphasis on developing winter malting barleys, resistance to BCOA could become more vital to malting barley production. Resistance to BCOA would protect malting barley from feeding damage, reduce BYDV incidence by a reduction in phloem feeding, and reduce the negative impact on winter hardiness. Greenhouse seedling screening to identify BCOA resistance was considered impossible because seedlings under standard conditions appear asymptomatic. A screening technique has been developed by this program in which seedlings show a varied response to BCOA feeding. Once this technique has proven valid for detection of resistance, screening of the entire NSGC of barley will begin.

SA has been reported to feed on more than 30 different species of grasses and cereal crops including corn and sorghum but wheat and barley are the preferred hosts. SA favors warm dry areas and feeding results in yellowing and withering of leaves which reduces crop yields. Early spring spraying is suggested for control but the aphid seems to prefer a more developed crop which may mean the necessity of a second spray later in the season. SA's ability to vector BYDV makes it a dual threat to barley yields. A field survey to determine the spread of SA is being supported by this project and should it be determined that SA has the potential to establish successfully in the US, screening of the NSGC for potential resistance will begin.

The most economical and environmentally sound solution to all these problems is resistant varieties. Negative effects on yield and malting quality are often associated with the use of identified unadapted resistant germplasm in a breeding program. A pre-breeding program will be initiated to bring aphid resistance genes from unadapted lines, identified as resistant from this program, into good malting quality backgrounds adapted to all the barley growing areas of the US. Due to the common occurrence in aphid populations of biotype change, after which the aphid can damage previously resistant lines, multiple resistant sources will be utilized in this pre-breeding program to produce genetically diverse resistant germplasm to protect barley from future aphid biotype change. A total of 58 adapted RWA-resistant germplasm lines have been released from this program with 36 in malting barley backgrounds. Since the introduction of RWA1 in 1986, 7 new biotypes (RWA2 – RWA8), have been identified. All 36 RWA-resistant adapted spring malting barley germplasm lines have been found to be resistant to RWA1- RWA5. Resistance to 4 other world biotypes has also been found in unadapted and adapted germplasm lines from this program. It is critical that we stay informed on future biotype changes and continue to test resistant lines to biotypes as soon as they are detected.

The second phase of the pre-breeding program is to determine the inheritance and genetic diversity of resistance in released lines. Inheritance studies give breeders information on how best to utilize these germplasm lines in their breeding programs. The genetic populations necessary for inheritance studies, to identify genetic markers, and to map resistance genes are being developed. Nineteen inheritance studies have been completed. Cooperative projects to identify genetic markers and map genes for RWA resistance have verified that genetic diversity for RWA resistance does exist in released USDA germplasm.

Marker Assisted Selection for Malting Barley Improvement

Kevin Murphy, Washington State University

The overall goal of this project is to use marker-assisted selection (MAS) to facilitate the selection of malting barley breeding lines in the WSU two-row spring barley breeding program. Using MAS will 1) allow for more efficient tests for the presence/absence of relevant traits; 2) evaluate a larger number of genotypes in earlier generations; and 3) enable selection during off-season cycles, thus streamlining the breeding process and increasing the rate of genetic gain. Towards this end, our goal is to identify predictive single nucleotide polymorphic (SNP) markers through genetic association analysis.

Malting Barley Production in Texas

Clark Neely, Texas A&M University

At one point in time, barley was grown on nearly 600,000 acres in Texas (USDA-NASS, 2014), but acres have steadily decreased since then to under 20,000 acres by 1999 (final year reported). As acres decreased in the state, so did active barley breeding and research. The last variety released by

Texas A&M AgriLife was ‘TAMbar 501’ in 2004 (Marshall et al., 2003). Recent interest by producers and consumers in locally grown ingredients has created a need for continued barley research in Texas. In fact, barley acres in Texas have seen a four-fold increase in acreage since 2009 according to FSA reported acres (USDA-FSA, 2015). Texas has not traditionally been considered a malting barley region; however, the development of a local malting facility and interest by in-state breweries for local ingredients provides a possible market. Ongoing malting barley variety trials indicates acceptable yield and malting grade barley can be produced in Texas, but the consistency of these results is still uncertain.

In the absence of a barley breeding program in Texas, 834 barley lines from the Triticeae Coordinated Agriculture Project were planted in headrows (single 3 ft row) in the fall of 2013 to test barley lines under multiple environments in Central and South Texas. This repository of barley germplasm is composed of genetic material from breeding programs across the nation and provides an opportunity to identify better adapted germplasm for Texas after appropriate screening is conducted.

The primary and long-term goal of this project is to screen for and identify high-yielding barley germplasm adapted for Texas environments that can be co-released with the original breeding program as one or more certified varieties marketed for Texas.

NATIONAL AND BASIC RESEARCH

Malting Quality of New Barley Selections

Stanley Duke, University of Wisconsin, Madison, WI

The Malting Quality Analysis project at Cereal Crops Research Unit in Madison has an annual mission of the timely return of accurate malting quality data of barley submissions to public sector barley breeders. We strive to maximize the return of these phenotypic results prior to spring and winter barley planting deadlines. This partnership allows the researchers to select and advance promising lines with the release of improved malting barley varieties as the ultimate goal. Barley subsamples are received from cooperating breeders; they are characterized, micro-malted, and the subsequent malts are analyzed for quality using several ASBC methods. The data are tabulated and returned to our cooperators who can then assess the suitability of the lines for development of commercial malting varieties.

A second major objective is the efficient support of AMBA’s testing program through barley characterization, malting, and analysis of Pilot Nursery (Midwest, West, and Winter), Mississippi Valley Barley Nursery (MVBN), and Western Regional Spring Barley Nursery (WRSBN) samples. We aim to provide complete data on these samples, ahead of the AMBA Spring Technical Committee Meeting. Another goal is to support biochemical and genetic investigations on barley and malt, by supplying raw materials (barley and malt) to interested researchers, and a final aim is the promotion of malting barley acreage through support to interested citizens, such as cooperative extension agents and growers.

We are exploring methods to increase sample throughput capacity, assuming adequate funding/staffing levels. This could improve our support of national barley researchers by increasing the amount of malting quality phenotyping available, annually. Presently, the limiting step in our processes is malting output. Weekly totals could be increased, up to double, by using half-sized malting containers, and malting smaller samples than our current 170g (d.b.) scale. A preliminary test yielded promising results, and once prototype half-sized germination cans are fabricated, we will

conduct a rigorous comparison of “reduced-sized” micro-malting (i.e. 100g d.b.) versus full-sized with our traditional malting system.

Temporal Expression of Ubiquitous Beta-Amylase During Mashing

Marcus Vinje, USDA-ARS, Madison, WI

The main objective of this proposed project is to determine if ‘ubiquitous’ β -amylase is expressed *de novo* during germination and malting. β -Amylase is one of the four main starch-degrading enzymes in barley malt and of these four enzymes correlates best with diastatic power. The majority of barley β -amylase research has been focused on the ‘endosperm-specific’ β -amylase but ‘ubiquitous’ β -amylase is more stable than ‘endosperm-specific’ β -amylase at higher temperatures, which is important due to the high temperatures involved during mashing. If ‘ubiquitous’ β -amylase is determined to be expressed *de novo* during germination and malting then this could give breeders and other researchers a new target for increasing β -amylase activity and subsequently diastatic power. ‘Endosperm-specific’ β -amylase is expressed and synthesized in the developing grains similar to seed storage proteins and it is also significantly and positively correlated with total protein. This relationship can lead to barley with high diastatic power and protein levels that do not meet malting quality standards. Increasing the ‘ubiquitous’ form of β -amylase in malt could circumvent the relationship between ‘endosperm-specific’ β -amylase and protein levels potentially leading to malting barley with high diastatic power that more consistently meets AMBA recommended protein levels. Additionally, more ‘ubiquitous’ β -amylase in malt could potentially lead to more efficient maltose production during mashing due to the ability to withstand common mashing temperatures better than ‘endosperm-specific’ β -amylase. Preliminary unpublished data on kilned green malt show an increase in ‘ubiquitous’ β -amylase between days 2 and 3 of germination indicating the potential for ‘ubiquitous’ β -amylase to be expressed *de novo* during germination and malting. The temporal ‘ubiquitous’ β -amylase expression pattern will be determined by tracking the transcript and protein levels using five biological repetitions of the malting cultivar Legacy grown in multiple environments from different crop years. These samples have already been micromalted and collected. This project is fairly straightforward with limited obstacles to overcome. Completion of this project should be accomplished within the funding year and published soon thereafter.

Investigations on Barley Diseases and Development of Low Temperature Tolerant Barley

Brian Steffenson, University of Minnesota

Plant diseases are one of the most important constraints to barley (*Hordeum vulgare*) production and quality in the United States. Our Cereal Disease Resistance and Germplasm Enhancement Project (CDRGEP) is part of the Minnesota Barley Improvement team led by Kevin Smith that develops two-rowed and six-rowed malting barley cultivars for the Midwestern United States and beyond. However, we also conduct disease evaluations for other barley breeders (both spring and winter programs) across the country. The primary mission of CDRGEP is the control of economically important barley diseases and development of germplasm with improved traits. The first part of this mission is best achieved through the development of cultivars with genetic resistance. Thus, the long-term goal of this research is to develop the knowledge base, resources, and germplasm for achieving durable disease resistance in malting barley cultivars. In conjunction with this goal, it is also essential to conduct barley disease surveys and monitor pathogen populations for new virulence types.

With respect to the second part of our mission for developing germplasm with improved traits, we are currently focusing on low temperature tolerance (LTT) in wild and landrace barley with the

ultimate goal of producing viable facultative/winter barley cultivars for the Upper Midwest and other regions. Our specific objectives for 2015/16 are to: 1) evaluate breeding lines for resistance to important diseases; 2) survey commercial barley fields for diseases; 3) collect pathogen isolates from barley cultivars for virulence determinations; 4) increase, maintain, and distribute pathogen stocks for testing barley germplasm for resistance and also barley stocks for testing pathogens for virulence; 5) collaborate with other investigators conducting research on malting barley and barley diseases; and 6) develop germplasm with LTT that can feed into the main barley breeding program. Our research goals all directly address AMBA's mission "to encourage and support an adequate supply of high quality malting barley for the malting, brewing, distilling, and food industries and increase our understanding of malting barley.

Our specific research objectives on disease resistance and LTT will increase barley competitiveness with other crops and support the development of higher yielding public and private sector malting barley varieties with characteristics that mitigate production risk factors to increase acceptance rates. All of the objectives listed above can be completed within a one-year period, with the exception of LTT parental line development, which will take three years. We anticipate no obstacles to fulfilling these goals.

Doubled Haploids and Foundational Winter Hardy Two-Row Germplasm

Patrick Hayes, Oregon State University

The goal of this research is to assist in realizing the potential of facultative/winter 2-row malting barley, an AMBA priority of national scope. Principal benefits are high yield and flexibility. Fall-sown facultative/winter types have high yield potential, typically 30% greater than spring-sown types. Facultative types have flexibility in planting date and thus reduce risk to growers and maximize continuity to maltsters and brewers. Doubled haploids are a powerful tool for barley variety development and genetics. They are particularly powerful for accelerating the development of winter barleys, where a vernalization requirement can delay generation advance through more traditional systems, such as single seed descent and/or off-season nurseries.

The OSU barley project has developed a doubled haploid production facility that has produced AMBA Pilot Scale candidates for the OSU program and germplasm for other U.S. programs. In this proposal, we seek to establish a foundation for a national collaborative program in winter hardy facultative/winter barley variety development by producing doubled haploids from crosses where one of the parents is of interest to participating breeders. The other parents will be the most winter hardy varieties identified in the TCAP LTT GWAS panel. The benefits will be i) a stream of winter hardy 2-row malting barley variety candidates, (ii) the foundation for a national collaborative GWAS and Genomic Selection project, and (iii) assurance of continued and increasingly efficient operation of the OSU doubled haploid lab.

Winterhardy Malting Barley Germplasm Development

Stephen Baenziger, University of Nebraska

The purpose of this research is to develop the most winterhardy winter barley germplasm in North America and to make it freely available to the U.S. barley community. As winter barley is becoming more important for malting and distilling, a second purpose is to increase the allele frequency of malting barley genes in our adapted winter barley so that others working on winter malting barley will have better parental germplasm for their programs. Two-row winter barley breeding will be emphasized as per the needs of the malting industry. However, barley in the Great Plains is also used for feed and forage and currently most of germplasm is 6-row, we expect some of the

developed germplasm might be 6-row. However, only the development of 2-row germplasm will be supported by this grant.

The key aspect of this research will be the use of testing sites in Nebraska which have previously been shown to reliably select for winterhardy winter barley, wheat, and triticale. These locations are unique in that they consistently stress winter barley so that winter tender lines are lost, but are not so severe that every line is winterkilled. For over 80 years, Nebraska has developed these sites and successfully created germplasm that has been used and studied by those most interested in developing winterhardy barley lines.

This research by its nature is long-term. However, for the upcoming year we expect to continue our breeding effort to enhance our selection of lines with superior winterhardiness and malting quality. We will plant our F₂ bulks at Lincoln, NE (a forgiving site for winter survival) to remove very winter tender lines and then plant the harvested F₃ seed in bulks at Lincoln, NE and Ithaca, NE. We will grow our F₆ and later generation experimental lines at Ithaca, Lincoln, and Sidney, NE. Ithaca NE is the site where we select for wheat winterhardiness and our hope will be to develop winter barley with winterhardiness similar to winter wheat or at least winter triticale which are competitive small grains in the Great Plains. Our main limitation will be variable climate which may affect our ability to selection. To enhance the frequency of malting barley alleles, we will increase our number of crosses to winter and spring malting barley types with an emphasis on 2-row malting barley parents. We are actively accessing the best global sources of 2-row winter malting barley through the appropriate and necessary material transfer agreements.

Metabolites Contributing to Malting Quality in Two-Row Barley

Adam Heuberger and Jessica Prenni, Colorado State University

The process of evaluating barley lines for malting quality is lengthy and expensive. Our previous studies (funded by AMBA) characterized metabolites from barley grain extracts using a mass spectrometry (MS) detection workflow and found that the metabolite profiles are correlated to multiple agronomic and malting quality traits [1]. This approach addresses major goals of the malting barley and brewing industry by enabling the (i) acceleration of the evaluation of barley lines for malting quality and (ii) detection of if and how growing location affects the overall profile of the malt, for a specific variety or breeding population.

Our previously funded AMBA research included a goal to establish a Barley Metabolome Library (BML) by acquiring UPLC-MS data on a diverse set of 192 malting barley lines. The goal of the BML project is to annotate all chemical signals observed in the mass spectrometer using non-targeted metabolite profiling of barley. For this study, our cooperators at AB-Inbev grew the population and measured malting quality. We recently completed metabolite profiling of 192 barley lines grown at 2 locations (referred to as the “AB-Global” population), and the data corresponded to an estimated 600 barley metabolites. We are currently comparing the UPLC-MS data to internal and external metabolite databases to annotate these compounds.

Our laboratory recently developed a new metabolite extraction procedure that allows for analysis using a second MS platform: gas chromatography-MS (GC-MS). GC-MS metabolomics is used to quantify metabolites that cannot be observed on an UPLC-MS system. These include some amino acids, sugars (mono-, di-, and disaccharides), and organic acids (e.g. citrate). Our initial barley research attempted to utilize GC-MS metabolomics, however high beta-glucan content interfered with the GC-MS acquisition. Our new extraction procedure reduces the oligosaccharide content of the metabolite extract and our preliminary results show that the extracts are now amenable to GC-

MS metabolomics. We believe that GC-MS metabolomics is critical to the development the Barley Metabolome Library because many of the compounds detected by this platform are known directly affect malting quality.

We recently prepared new metabolite extracts from the 384 barley samples using the modified extraction procedure. Therefore the overall objectives of the proposed work are to: perform GC-MS metabolomics on the newly prepared AB-Global barley grain extracts and incorporate GC-MS detected metabolites with the UPLC-MS Barley Metabolome Library

We anticipate no significant obstacles with the completion of the proposed work and these goals can be met within the 1 year funding timeframe. Preliminary work has already been completed to establish an effective extraction method for barley. Furthermore, our laboratory has an established a routine GC-MS metabolomics workflow for data acquisition and analysis that results in the detection and annotation of ~50-200 metabolites, depending on the sample. This study directly addresses the AMBA strategic goals to “develop technology to accelerate variety development” and to “improve quality evaluation for breeding programs” and “fermentability prediction”. Our previously funded AMBA studies provided the groundwork for incorporating metabolite markers into malting barley breeding programs. Future efforts could determine if GC-MS detected malt metabolites correlate with variation in barley metabolites, investigate the dynamics of GC-MS detected compounds with malting.

Bacterial Leaf Streak of Barley

Zhaohui Liu, North Dakota State University

Bacterial leaf streak (BLS), caused by *X. translucens* pv. *translucens* (*Xtt*), is the most common bacterial disease of barley. Recently, BLS has reemerged as an important disease in the North Dakota and surrounding areas where the majority of malting barley is produced in the United States. The disease can cause significant yield losses and grain quality reductions. No chemical and other cultural practices can be used to control the BLS in the field, and thus using resistant cultivars appears to be the only option. The current cultivars grown in this region are likely susceptible based on our observations in the field, but the accurate information on their reactions is not known. Furthermore, source of BLS resistance for breeding programs is lacking. Our goal is to determine the level of resistance/susceptibility in the current cultivars, identify sources of resistance, and map the resistance genes/QTL. This research will provide valuable information and tools for the development of BLS resistant malting barley cultivars, which in turn reduce the risk of inadequate supply of high quality of malting barley due to BLS epidemics.

Genetic Analyses of Winter Hardiness & Malting Quality

Erick Stockinger, Ohio State University

The long-term goals of the project are to develop two-row winter malting barley varieties adapted to Ohio and our neighboring Great Lakes, Midwestern, and Northeastern State regions. Another goal is to develop genetic resources that enhance our understanding of winter-hardiness and malting quality at the genetic level. The activities associated with attaining these goals are intended to be complementary. The specific objectives during the one year funding period are to advance eight genetic populations at least two generation cycles in the greenhouse. These populations are derived from crosses between a panel of different winter-hardy two-row lines and one two-row line that lacks winter-hardiness but has high malting quality. A longer-term goal associated with development of these populations is to use them to better understand the genetic basis of winter-hardiness and malting quality. The plan is to advance the populations from the F₃ generation to the F₅ generation,

and possibly the F_6 by autumn 2016, when we plan to begin field testing. Characterization of freezing tolerance, winter-hardiness, and malting quality and subsequent genetic analyses of these traits is anticipated to require a three to five year timeframe. Similarly were new varieties to arise from these efforts, release of that new variety would have a timeframe of five or more years. While the target is to develop 125 RIL individuals in each population, it is anticipated that one issue will be natural attrition of some individuals in each population. If certain populations fall below numbers required for genetic analyses we will carry out the genetic analyses with only those populations having the required number of individuals while still carrying forward the populations having too few individuals in the event that one or more lines in those populations may exhibit superior agronomic performance in the field. Together these activities are anticipated to create a foundation base of germplasm that can be used in subsequent crosses with other malting barley lines and exchanged with other barley breeders, enhancing winter malting barley breeding and germplasm development in the public domain.

Flavor profiling of two and six-rowed winter malt barley varieties for Virginia

Sean F. O'Keefe and Brian Wiersema, Virginia Tech

There is a demand for brewing malt from local-grown barley in the Eastern United States. Virginia Tech has initiated a breeding program to support this demand, but we have not had the tools available to malt barley and evaluate its flavor quality until very recently. The Food Science Department has recently acquired a 2.5HL brewhouse and pilot malting equipment, opening the door for our preparing and evaluating malt from our breeding lines. This research will initiate our work on malting and flavor profiling of malted barley from winter malt varieties from the Virginia Tech breeding program.