

## **BREEDING STRATEGIES FOR BARLEY QUALITY IMPROVEMENT AND WIDE ADAPTATION**

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ABSTRACT. Barley is one of the five major crop species of the world which widely use for stock feed, human food, malting, brewing and distilling. Barley provided familiar source of animal feed and food as well bear which is popular and traditional drink. This plant species represents low protein crop and it is very important for malt quality and brewing development. The region with low rainfall and low soil fertility combined with local crop rotational requirements provides conditions to produce optimum malting quality in different barley cultivars. A wide range of variables, including abiotic and biotic stresses can affect quality components. While each users of barley require consistent raw material, economic price and specific quality, the barley breeders need to create new barley cultivars. Initial breeding and selection objectives were focused on agronomic trait as grain morphology, yield and disease resistance. Crop management play important role in barley spreading and yield improvement. Breeders need to develop cultivars for markets which demand clean bright grain with low moisture, better disease resistance, and high quality parameters. Significant consideration when breeding must be given barley cultivar ability to achieve quality in different environmental conditions.

### INTRODUCTION

The barley breeding program represents the characterization of agronomic, grain and malt quality attributes of potential new cultivars. Testing of newly cultivars in different environmental condition giving us possibility of estimation genotypes/environments interactions and making complex of genotype characterization. Grain developments influenced by all aspects of the production environment and the biochemical complexity of ‘malting quality’ and making difficulties of defining a genotype’s quality attributes. Weather conditions represents a major

source of environmental variation as well as soil type, fertility and moisture status of grain development. It is clear that we could be get advantage to evaluate comparable grain samples of genotypes which have been grown under similar conditions. This would allow the identification of relationships between grain protein content and other quality parameters. Development of new spectroscopic and image analysis tools have enabled the development of fast multivariate screening analysis on the level of phenotype. Different brewing practices can lead to very different quality specifications. Tradition of brewing for example preference for two-rowed cultivars or six rowed cultivar can lead to different definition of quality, too.

Studying the biochemistry and genetics malting quality focused on the systematic characterization of starch deposition and hydrolysis as well on controlling proteolysis. This research make clear biological processes but has not provided breeders with a better tool kit for improving malting quality. Breeders conducting expensive test for establishing the malting quality of experimental material including analysis of malting quality genetics and than malting quality phenotype which have shown frequency distribution according to Mendelian inheritance. Accordingly, breeders conducted phenotypic selecting for malting quality. These procedure have had limited by practical applications.

Quantitative trait locus (QTL) methods represent a step forward in applied genetics that providing assessment of numerous genes, their location and controls of phenotypic traits. In the case of malting quality, this genome location information allows for the testing of hypotheses about genes determining QTL and it provides tools for marker-assisted selection (Beavis, 1998; Hayes and Jones, 2000). Molecular marker analysis allows selection at the level of the genotype rather than the phenotype. This is useful for traits which are difficult to estimate in the field such as tolerance to various stresses (This et al., 2001).

Barley (*Hordeum vulgare* L.) ranks as one of the most widely adapted plant species grown in near-arctic and semi-desert environments. The initial study of wide adaptation in barley based on data from international nurseries. Yield trials from 12 International Barley Yield Nurseries were used for identification of barley crosses with good performance. Data from 44 locations were classified into three groups: eight top-yielding lines, eight intermediate types and eight lowest-yielding lines. These 24 Mexican lines were sown with the best adapted local cultivars as the check. The line derived from the cross LB Irna/UNA8271//Gloria/Come belongs to the highest yielding group at 25 locations, to the middle group at 11 locations and to the lowest yielding group at 8 location. The similar results had check cultivar. In contrary another cultivar Tamina was classified in the highest yielding group at 11 locations while in the lowest yielding group at the 22 location. These data indicted that lines expressed wide adaptation while Tamina cultivar the narrow adaptation (Vivar, 1996).

According to the Consultative Group on International Agriculture 1.4 bilion people are dependent on agriculture in arid regions where scarcity of water not land is the problem. Arid and semi-arid regions account for approximately 40% of the world land area. Drought is perennial problem particularly in Asia, the Middle East Africa. Also, climate change now seriously affects agricultural productions in cereal production areas in Europe and elshewere (Forster et al., 2000). Barley is use from livestock feed to specialized food staffs and particularly well known use is for the production of malt used in brewing and distilling. This plant species is common crop in drought area where inputs are low and where there is a high risk of crop failure because drought causes decreasing not only yield but also grain quality. Pragmatic strategy in breeding barley for stressed environments have been conducted by International Center for Agricultural Research in the Dry Areas. Their strategy based on decentralization of breeding to exploit locally adapted

germplasm, assessing correct germplasm in target environments, selection in farmers fields, the use of heterogeneous seed (mixtures and landraces) Ceccarelli et. al., (1996).

The main goal of barley breeding is improvement of stress tolerance that represents a major goal for the plant breeders and for the agriculture in the future. Necessity of developing sustainable agriculture in arid condition in the times of global climate changes indicated complexity of this task. Also, stresses can be caused by biotic and abiotic factors which may influence to the strategy of breeding and selection of wide adopted cultivars or selection for specific (favorable or unfavorable) environment.

## GENETICS OF DROUGHT TOLERANCE

The adaptation of barley plants to environmental conditions has been studied for many years. Genetic studies have focused on selection of cultivars which are more tolerant to environmental stress caused by different environmental conditions as well cultivars with improved quality and increased yield and yield stability. Abiotic stress tolerance mainly controlled by many genes.

Investigation of drought tolerance has been focused and developed in collaboration between physiologists and breeders (Blum et al., 1994, Ceccarelli). Study of numerous physiological and biochemical traits (leaf turgor, osmotic and photosynthetic adjustment, relative water content, water use efficiency, abscisic acid content, stomatal conductance, transpiration efficiency, mobilization of soluble stem carbohydrates and senescence) as well study of developmental, morphological and ultra-structural traits (leaf emergence, leaf area index, leaf waxiness, stomatal density, tiller development, flowering time, maturity rate, cell membrane stability, yield and yield components) lead to the concept of pyramiding physiological traits (Yeo et al., 1990) and definition of ideotypes for drought conditions.

So, for environments with mild winters favored ideotypes need characterized early heading, early vigor, erectile winter growth habit, tolerance to cold damage while for harsh winter conditions favored ideotype expressed medium-early heading time, prostrate winter growth habit, good cold tolerance. The ideotypes may reflect variation for vernalisation and photoperiodic responses (Forster et al., 2000). Collaboration of physiologists and breeders represents contribution to rapid and effective screening that physiological traits could be integrated into breeding programs. Plant architecture can be an important marker of determining of water stress resistance. Also root characters play an important role in soil exploration for water. Tiller development and leaf growth are important morphological traits response to drought that were studied in barley (Teulat et al., 1997). A QTL controlling several growth parameters was co-located with a QTL controlling relative water content.

However, drought tolerance traits identified by physiology are often genetically complex and selection for these has not succeeded under drought conditions. An example of difficulty in translating physiological study into genetic mechanism is root growth. Recent advances in genetics, increasing knowledge in genetic markers, genetic maps and quantitative trait loci (QTL) analysis, have been exploited for this aim. These genetically mapped can be tested specific physiological trait/responses and thereby detect the number of genetic loci involved in individual and combined effects. Physiologists can provide understanding of phenotypes corresponding to certain genotypes. The genetics and barley breeders need integrated into new genotypes. Abscisic acid (ABA) plays important role as a stress response signal. It has been studied extensively in

barley and other cereal species (Quarrie et al, 1997). Drought related traits studied by QTL analysis include: content of leaf ABA, relative water content and osmotic adjustment in barley (Quarrie et. al., 1994; Teulat et al., 1998). However the locations of QTL for leaf ABA content are not always conserved when estimated over several years. The xylemic ABA have a major regulatory effect on stomatal conductance. Water use efficiency (WUE) can be assessed by carbon isotope evaluation in grain or leaf. Several region of barley chromosome associated with genetic control of WUE. A major region on chromosome 4 was found for carbon isotope discrimination of shoot measured from salt stressed plants (Ellis et al., 1997). Several regions involved in osmotic adjustment parameters (Teulat et al., 1998).

The QTL approach provides markers for areas of the genome involved in stress tolerance that can coincide with yield and quality QTLs or other important genomic regions (Foster et al., 2000). The genetic markers can be used for the number of purposes: a) developing gene map location against QTLs for drought tolerance, b) screening germplasm for variation c) predicting phenotypic responses to drought tolerance d) using in marker assisting breeding. The development of molecular markers and genetic maps has allowed the identification of markers linked to genes economically important traits which can be exploited in plant breeding (This et al., 2001). Marker-assisted selection for those traits would than be of great interest. Genetic control of drought resistance is very complex and QTL identified for a range of drought response traits show variation with the environment and genetic background. To identify QTL controlling yield and its components in drought stress condition will be very helpful.

Molecular markers can provide identification of genes response to winterhardiness, resistance to salinity stress and their chromosomal location in barley. Winterhardiness tolerance QTL were identified on chromosomes 5 and 7 (Oziel et al., 1996; Tuberosa et al., 1997). Large number of genes may be involved in the drought, salinity and cold tolerance. A lot of genes have been isolated in abiotic stress while some of them may be candidate genes for drought controlling. It will be worthwhile to establish which of these stress-induced genes map at QTL for related stress resistance traits. Therefore, many stress induced genes may have small effect on improving stress tolerance.

## BREEDING BARLEY FOR MALT QUALITY

Breeders need to develop new malting cultivars with better malting and feed quality. Barley for malting is required to be of low protein content and high starch content. Very often quest for yield improvement can create barley less well suited to the end uses of malting. Numerous cropping practices may influence to grain protein concentration by high rate of nitrogen application, late sowing and sowing on low moisture. The negative correlation between malt extract yield and protein – mainly due to hordeins, the major fraction of endosperm storage proteins, is well known. Some investigation indicated that this negative correlation can be influenced by genotype x environment interaction (Maksimovic et al., 1994; MacLeod, 2000).

Malting and brewing have been in use for more than 6000 years. Different phase of process known: manual handling, pneumatic malting and automation and refrigeration process. The major quality issue at each phase of process was associated with grain size, low dormancy and uniform germination because it is important for homogeneous malt product. On the malt homogeneity have influence endosperm density, impact of starch and protein, cell wall thickness, husk content as well hormone and enzyme synthesis. All these traits are considered important

targets in breeding new malting barley cultivars. The major goal of breeders need to create malting barley for brewing with maximum extract yield, sufficient nutrient for yeast growth, fermentable sugars for alcohol production and balanced combination of high molecular weight compounds to contribute to mouth feel and foam and flavor quality.

For brewery is very important that malting barley cultivars need to be with low protein content and high starch content to maximize the potential for extract yield. Extract is very important for alcohol production. However protein level can not be too low because protein serves four basic functions in brewing. It is origin of all enzymes that catalyzing the complex biochemistry involved in turning barley through malt into wort. It is required for yeast nutrition. It contributes to foam what is involved in formation of flavors that malt contribute to beer. Understanding of the complex biochemical and chemical pathways occurring during the malting of barley that set of aims for new malting barley cultivars. Having developed the technology to measure a particular character the next challenge is establish whether that characteristic can be controlled or manipulated in the barley in the breeding program or in the malt (by maltster) or both to optimize brewing process.

Malting quality is a complex character. By traditional breeding the most agronomic traits can be tested or visually scored during the growing period. The development of assessment techniques have led to gains in breeding productivity. Numerous individual traits controlling by number of genes and traits can be linked to other less desirable. Also agronomic and malting quality traits are quantitative and environmental factors can influence to the level of their expression. Recent biotechnology studies have mapped genes responsible for many agronomic and malting quality traits and increase knowledge of gene structure and function. Molecular marker technology is now used in many breeding programs, which allows rapidly selection of large number of traits and in single generation. Also, use of double haploid techniques to fix a selected genotype can also reduce number of years required to develop improved cultivar.

By using modern techniques stable barley transformation was presented. It means that genetically modified malting barley and technology gives the ability to enhance specific quality traits. The first aim for genetic manipulation was increasing yield and improving production practices. Genetic modification of malting barley has potential for benefits to producers, processors and consumers through adding or removing specific quality characteristic. However, it is necessary to identify traits which will be specific direct relevant to the final consumer or beer drinker, because of suspicion based on lack balance the perceived risk and benefits of the technology. Development of modified malting barley cultivars and their commercialization made changes on global market, public and private companies and government activity on development of legislative for using, trade and labeling goods and products from genetically modified cultivars.

## FUTURE OF BARLEY GENETICS

Barley has been useful genetic experimental organism which attributable with its diploid nature ( $2n=2x=14$ ) self fertility, large chromosome (6-8  $\mu\text{m}$ ) high degree of natural and easily inducible variation, ease of hybridization, ease of doubled haploid production and wide adaptation. These characteristics are valuable for many classical genetics studies such as cytogenetics and mutagenesis. Traits of barley's diploid nature, ease of hybridization and ease of

doubled haploid production are important traits that make it a highly suitable for molecular study and Quantitative Trait Loci (QTL) mapping.

Barley genetics have made important contributions to the field in both practical and theoretical terms. The genetics is undergoing a major shift from “one gene at a time” to a genomic approach which promises to lead to an understanding of all of the genes that make up an organism leading to potential reshuffling of the genome for new knowledge and new cultivars at a rate that has not been possible by evolution or traditional plant breeding (Kleinhofs, 2000). The genomics investigation represents an intensive and expensive increasing the necessity to focus model organisms and model systems. Barley geneticists will adapt to this situation by exploiting the small genome model organisms like rice, applying the barley genome model to more cumbersome genetic organisms such as wheat, and focusing their genomics efforts on a few highly developed systems. However barley will not displace rice as the model grass genome but it will make important contributions as a model *Tritice* genome, and to functional aspects that are unique to barley. Barley genetics future is bright and will important genetic systems among the plants.

Recent technological developments such as micro arrays and identification of gene rich regions make genome size less of an issue than previously believed. Nevertheless, the gene functionality identification is resolved and transformation technology becomes readily and reliability available it will become possible to insert from any species into economic crop plants. It could be big contribution to barley characterization and germplasm collection. The process of describing genetic variability at the molecular level is almost trivial, but the mechanism for the generating and maintaining DNA polymorphism are complex (Powell and Russel, 2000). Variability of Barley gene pools provides opportunities to expend the profitability of barley breeding and productivity. Numerous studies reported the spectrum of variability in various barley gene pools but it is necessary identify significance of this variation and how we can best exploit this knowledge and improve barley breeding programs. Developments in genome science contributed us to recognize the importance of connecting genetic and physical maps to phenotypic data. Mapping of barley genome giving us data of gene location controlling complex phenotypes. The aim of many barley research programs is to identify genes controlling yield and quality by QTL mapping. By developing series of isolines, in which small segments of a donor genome are introgressed into a common recipient providing a library of lines covering the whole genome, will allow improved QTL identification. Monitoring the effects of selection during the breeding process in numerous genetic backgrounds will also allow us to cost effectively utilize a large amount of data that has been reported during barley cultivar improvement and enable the targeted utilization of germplasm resources.

## BREEDING FOR DISEASE RESISTANCE

The modern agricultural practices and continuous genetic improvements grain yield has tripled within the past century and continuous to make a new progress. The breeders have led to creation of cultivars adopted for diverse environments on all continents and for different purpose as feed and brewing. With yield increasing, the genetic variability on barley fields gradually decreased because local populations and landraces were replaced by cultivars. The analysis of both pedigrees and molecular data shows that many of these cultivars share related pedigrees (Graner et al., 2000). The genetic uniformity of today barley fields makes possibility for the

epidemic spread of any disease. Breeding for disease resistance is a major goal in any breeding programs and consists two steps, first – identification of donor resistance and second – transferring of the corresponding genes into a breeding line. The potential of molecular marker technologies for barley breeding as well as for genetic studies is in dependence of numerous agronomics important traits that have been tagged. The process ranging from the identification of appropriate resistance donors to the development of a selectable marker requires collaboration of barley breeders, phytopathologists and geneticist.

Considerable progress has been achieved regarding the identification of major genes conferring resistance to the leaf rust pathogen *Puccinia hordei*. Molecular mapping of the race specific genes *Rph2*, *Rph12* and *Rph9* as well genes *Rph7* and *rph16* make clear that there are two genes for *Puccinia hordei* pathotypes and that their combination in one genotypes might represent an efficient way to increase plant resistance. The co-dominant PCR-markers were developed for *Rph7* and *rph16*, permitting a marker assisted combination without any progeny testing. The similar is with resistance to stem rust caused by *Puccinia graminis*. The *rpg4* gene confers complete resistance and can be replaced or combined with the dominant *Rpg1*. Regarding to the net blotch disease, a *Rpg1* gene conferring resistance to selected pathotypes of *Pyrenophora teres* has been located on chromosome 2HS (Graner et al., 2000). In addition resistance to fungal and viral pathogens there are two genes conferring to the cereal cyst nematode *Heterodera avenae*, which have been located on chromosome 2H and 5H (Barr, 1998). Also, gene *yd2* merely confers tolerance to barley yellow dwarf virus (BYDV) located at the proximal region of 3HL chromosome where located additional resistance genes (*Rhy*) against *Rhynchosporium secalis* and *Pyrenophora teres*. Various strains of the BYDV have been identified and transmitted by different aphid species, while in the contrast to BYDV, barley yellow mosaic virus (BaYMV) and barley mild mosaic virus (BaMMV) are soil born pathogens wwhich are transmitted to the roots of seedlings by the fungal vector *Palymixa graminis*. Locus *rym4* in telomeric region of 3HL chromosome conferring immunity to BaMMV and BaYMV while second locus conferring resistance to virus complex located at the 4H which is allelic and described as *rym11* locus. Expect different biotic stesses, mainly artropods and diseases there are insect pests which limit production potential of barley. The barley stem gall midge, *Mayetiola hordei* Keiffer, Russan wheat aphid, *diuraphis noxia* Mordivilko has prevailed as a pest of barley in particular region of Europe, Africa and America.. For the barley breeders is very important identify source of resistance to insect pests and by breeding transfer gene resistance and develop resistant cultivars. By screening of wild barley collection were identified sources of resistance *Hordeum marinum* and *Hordeum bulbosum* that showed antibiosis to the fly (Lhaloui, et al., 2000).

Gene mapping is very important for the breeding program. Also identification of novel resistance genes and their isolation may lead to the development of diagnostic markers that can be used for a rapid differentiation of alleles in germplasm collections. Wild species closely related represent an important source of genetic resistance to pest and pathogens. Using of wild barley as a source for barley breeding we need to know what pathotype strategy to adopt (local, exotic, pure, mixed), whether resistance is prevalent, rare or absent, what is genetic bases of any resistance, whether wild resistance shows predictable geographic patterns, relationship between wild resistance barley cultivars, whether molecular tags available to expedite breeding etc.

## SUMMARY

The problem in barley breeding is the relationship between selection and target environment. Breeding of barley breeders can realize in two direction i.e. selection for broad and specific adaptation. Breeding for wide selection have conducted enough breeding cycles under optimal or suboptimal environments and cultivar selected for favorable and unfavorable environments. It means creation cultivars for wide adaptation over time and wide adaptation over space. Breeding for specific adaptation to unfavorable condition is often connected with undesirable breeding results, because it is usually associated with a reduction of quality or yield potential under favorable conditions. Considering this breeders need achieve aim to create cultivars that will maximize yield in favorable condition, too.

Traditional breeding method has been very successful in improving yield and quality. On the base of genetic variability were developed cultivars of barley by breeding process. Recently plant breeders use mainly adapted commercial cultivars for crossing. This way contributes to decreasing genetic variability. For increasing genetic variability it is necessary use wild species for crossing in barley breeding program. By such crossing we will able create genetic variability among the progeny which requires selection of combination with desirable traits and further crossing to fix the selected genotype.

QTL tools, by assessment of genome location, identification of number of genes that determine complex phenotypes have a significant contribution to improving of barley genetics and breeding. In the case of malting quality we can begin integrate knowledge of malting biochemistry with malting barley improvement. Genetic diversity for malting quality which exists in barley represent important source for selecting genotype with high adaptability on the production areas condition. QTL mapping is a useful tool for barley genetics and breeding especially for the estimation of economically important functions to specific regions of the genome. By tools for sequencing gene rich regions we will be able systematically differentiate between structure and function and partition end-point phenotypes into their regulatory and structural components.

The modern crop breeding technology, including transgenic material, has the potential to accelerate the breeding process and to focus specific quality characteristics. It also offers opportunity to manipulate quality for example it can deliver capability of improving the thermo stability of a particular enzyme or increasing the expression of specific enzymes. It has potential to add new enzymes achieve the radical change without disturbing the majority of cultivars characteristics. However, it will not doubt that breeders after use of biotechnology map will meet many of needs of the producers and processors of malting barley and will continue to use traditional breeding techniques.

The ability to select specific genes such as disease resistance has allowed pyramiding of several different sources of genes controlling resistance to multiple strains. The wild relatives (*H. bulbosum*, *H. murinum*) breeders need as a donor of genes encoding quality traits and disease resistance. The success in crossing of wild relatives of barley *Hordeum chilense* and durum wheat and development of new hexaploid crop plant *x Tritordeum* Ascherson et Graebner indicated that wider crossing program cold be open direction of identification novel useful hybrids.

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