Barley Genetics Newsletter

Volume 36

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Acknowledgements

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Barley Genetics Newsletter 36:1-2 (2006)

A new allele in a *uzu* gene encoding brassinosteroid receptor

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Brassinosteroids are growth-promoting natural products, found at low levels in pollen, seeds and young vegetative tissues throughout the plant kingdom. They play an essential role in diverse developmental processes including cell expansion, vascular differentiation, etiolation and reproductive development (Clouse and Sasse, 1998). It has been found that semidwarf barley accessions carrying the *uzu* gene are non-responding to brassinosteroids. The synteny suggested that the *uzu* gene may be homologous to rice *D61*, which is a homolog of *Arabidopsis thaliana BR-insensitive1* (*BRI1*), encoding a BR-receptor protein. A barley homolog of *BRI1*, *HvBRI1* (acc. no. AB088206) was isolated (Chono et al., 2003). The sequence analysis showed that uzu phenotype was caused by single-nucleotide substitution A > G at the position 2612 of the *HvBRI1* gene. This mutation resulted in an amino acid change at the highly conserved residue (His-857 to Arg-857) of the kinase domain of BRI1 receptor protein and caused the reduced sensitivity to BRs and reduced plant height (Chono et al., 2003). We have found a semidwarf mutant 093AR, produced by MNU (N-methyl-N-nitrosourea) treatment of variety Aramir, that proved to be allelic to the uzu form. The aim of this work was to determine and compare the sequences of *HvBRI1* gene between spontaneous mutant uzu, induced mutant 093AR and its parent Aramir variety in order to identify the putative mutation(s).

DNA extraction was performed according to “micro C-TAB” protocol (Doyle and Doyle, 1987). Six primer pairs were designed on the basis of *HvBRI1* sequence (GenBank acc. no. AB088206) utilizing the Primer3 program (http://frodo.wi.mit.edu/cgi-bin/primer3). PCR reactions were performed using the “touch-down” strategy. Three independent PCR reactions were performed for each primer pair and amplification products were extracted from agarose gels using QIAEX II Gel Extraction Kit (QIAGEN) and cloned into pGEM-T Easy vector (Promega). Plasmid purification was carried out using QIAprep Spin Miniprep Kit (QIAGEN). Inserts were sequenced using SequiTherm EXCEL II DNA Sequencing Kit (EPICENTRE) utilizing LI-COR IR2 sequencer.

We determined the sequences of *HvBRI1* gene in the genotype uzu, 093AR and Aramir variety. The single-nucleotide substitution has been confirmed at the position 2612 (A-2612 to G-2612) in the spontaneous mutant uzu, which proved to be specific for this genotype. This mutation is localized in highly conserved kinase domain of receptor protein. The comparison of sequences of *HvBRI1* gene between mutant 093AR and Aramir variety, from which this mutant was obtained, led to the identification of the C to A substitutions at the positions 1760 and 1761 in the mutant (Fig. 1).

1 cgcttctcgcatggtctcaaggtagagcgagcgagctcccacatggattg

1701 gatgccgagcaagaagctgtgcaacttcacaaggatgtacatggggagca

1751 cagagtata**cc**ttcaacaagaatggctccatgatatttctggatttgtca

1801 tttaatcagcttgactcggagatacccaaggagcttggcaatatgttcta

…

2551 gtgagtggccagggtgaccgggagttcacagcggaaatggagaccattgg

2601 caagatcaaac**a**ccgcaaccttgttccgctcctcggctactgcaagatcg

2651 gcgaggagcggctgctgatgtatgacttcatgaagtatggcagcttggag

…

3501 gcctctcagatgattgatgatgatgtgatgataccattctgaggttccca

3551 cgcaagct

Fig. 1. The sequence of *HvBRI1* gene (acc. no. AB088206). **cc**- at the positions 1760 and 1761 indicate C > A substitutions in the 093AR sequence; **a-** at the position 2612 indicates A > G substitution in spontaneous mutant uzu.

On the basis of *in silico* translation using Jellyfish program it was postulated that this mutations lead to the replacement Thr-573 by Lys-573 (Fig. 2).

1 mdclrlavaaaalllaalaaaaddaqlldefrmalpsqaplegwtarega

…

501 ippelaeqsgkmtvgliigrpyvylrndelssqcrgkgsllefssirsed

551 lsrmpskklcnftrmymgsteyt**t**fnkngsmifldlsfnqldseipkelgn

601 mfylmimnlghnllsgaiptelagakklavldlshnrlegqipssfssls

…

1101 pgfgvmdmtlkeakeekd

Fig. 2 The sequence of HvBRI1 protein (acc. no. BAD01654). **t** – indicates the Thr-573 to Lys-573 conversion, deduced by *in silico* translation, caused by C > A substitutions at the positions 1760 and 1761.

The threonine residue is localized between last LRR (Leucin-Rich Repeat) domain and pair of cysteines participating in protein dimerization, followed by transmembrane domain. It was shown utilizing ClustalW program that Thr-573 residue is highly conserved among BRI1 homologs from barley, rice, arabidopsis and tomato. As threonine and lysine have different chemical properties, their substitution within the conserved domain may lead to the change in protein activity.

References:

Chono, M., I. Honda, H. Zeniya, K. Yoneyama, D. Saisho, K. Takeda, S. Takatsuto, T. Hoshino and Y. Watanabe. 2003. A semidwarf phenotype of barley uzu results from a nucleotide substitution in the gene encoding a putative brassinosteroid receptor. Plant Physiology. 133:1209-1219

Clouse, S. D. and J. M. Sasse. 1998. Brassinosteroids: essential regulators of plant growth and development. Annu. Rev. Plant Physiol. Plant Mol. Biol. 49: 427-451.

Barley Genetics Newsletter 36:3-5 (2006)

Response of Induced Dwarf Mutants of Barley to Exogenous Application of Gibberellic Acid

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Abstract

Response of five induced plant height mutants (dwarfs and semi-dwarfs) of barley to gibberellic acid (GA_3) treatment was studied. The gibberellic acid response index (GRI) was calculated and based on this value, the mutants have been categorized as sensitive (high or moderate) or insensitive. The GRI value was highest for dwarf mutant followed by early maturing mutant with the former being highly sensitive to GA_3 treatment while the latter was moderately sensitive. On the other hand, semi-dwarf and chlorina mutants and the control were insensitive to exogenous application of GA_3.

Key words: GA_3 treatment, reduced height mutants, barley, gibberellic acid response index (GRI).

Introduction

Gibberellins stimulate extensive growth in intact plants. These enable to overcome genetic dwarfness in some species if that dwarfness is because of a gene mutation, resulting into blocked gibberellin production. Lack of gibberellins causes shortening of internodes and reduces the height of the plant. Exogenous application of gibberellic acid (GA_3) fulfils its requirement and the plant achieves normal height. Gibberellins have little effect when they are applied to plants of normal height. The most outstanding effect of gibberellins on plants is the abnormal stem elongation. This is brought about primarily by increased cell elongation, and secondarily by accelerated cell division. Under its influence generally dwarf varieties have been changed in to tall varieties in some species. The small size of genetically dwarf plants is due to some natural growth inhibitors. The rapid elongation of the stem that follows the application of gibberellins is attributed to the neutralization of such inhibitors by gibberellin.

The development of semi-dwarf cultivars has occurred with little understanding as to the effects of the plant height genes on plant traits other than straw length. In wheat, traits pleiotropic to or closely associated with, *Rht1* and *Rht2* genes include gibberellic acid insensitivity, cell size and cell number, root weight, coleoptile length, leaf size, grain yield, yield components, biomass, harvest index, protein content, and disease reaction (Gale and Youssefian 1985). In any mutation breeding programme, reduced height mutants are one of the most frequently occurring class of mutations. However, there are not many studies dealing with the response of dwarf mutants to GA_3 treatments. Hence, the present report on the response of induced dwarf mutants of barley to exogenous application of gibberellic acid.

Materials and Methods

A laboratory experiment involving five induced dwarf mutants of barley along with parental control was conducted at the department of Agricultural Botany, Ch.

Charan Singh University, Meerut during 2002-03. The M_6 seeds of the five true breeding barley mutants viz. dwarf, semi-dwarf, early maturing, semi-dwarf with early maturity, and chlorina types (isolated from gamma irradiated populations of barley cultivar 'K 169') were used. Response of reduced height mutants to GA_3 was estimated following Yamada (1990). For the GA_3 response test, seeds were germinated in petri-dishes for 2 days at 20^0C, then transferred to seed trays containing a mixture of vermiculite, soil and sand in the ratio of 2:2:1 by volume. The seedlings were grown in trays in a growth chamber under controlled light and temperature conditions. After 5 days of growth at 20/15 ^{0}C (day/night) for 16/8 hours, the seedlings were supplied with 10 ppm GA_3 through irrigation water every other day. Seedling length and leaf sheath measurements were recorded on 20 days old seedlings.

Results and Discussion

The data recorded on seedling length and the length of first leaf sheath of 20 days old seedlings of both GA_3 treated and untreated mutants and control are presented in table 1. The gibberellic acid response index (GRI) was calculated and based on this value, the mutants have been categorized as sensitive (high or moderate) or insensitive. The GRI value was highest for dwarf mutant (141.81) followed by early maturing mutant (113.18). The dwarf mutant was highly sensitive to GA_3 treatment while the early maturing mutant was moderately sensitive. A moderate response was also recorded for the semi-dwarf mutant with early maturity while the other two mutants of the present study and the parental control were insensitive to GA_3 treatment (Table 1).

Table 1. Seedling and leaf sheath measurements (mean ± S E), GA_3 response index (GRI), and GA reaction in gibberellic acid treated seedlings of reduced height mutants in barley.

Control/Mutant	Length of first leaf sheath in		GRI (%)	Reaction to GA_3	Seedling length in		GRI (%)
	GA_3 treated	Un-treated			GA_3 treated	Un-treated	
'K 169' control	18.25 ± 0.34*	17.85 ± 0.21	102.34	Insensitive	23.16 ± 0.32	22.45 ± 0.34	103.16
Dwarf	11.70 ± 0.66	8.25 ± 0.21	141.81 **	Highly sensitive	13.66 ± 0.16	10.32 ± 0.74	132.36**
Semi-dwarf	13.33 ± 0.67	13.10 ± 0.48	101.75	Insensitive	18.20 ± 0.46	17.84 ± 0.58	102.01
Semi-dwarf w/ early maturity	13.05 ± 0.48	12.37 ± 0.50	105.49	Mildly sensitive	17.42 ± 0.36	16.32 ± 0.51	106.74
Early maturing	15.28 ± 0.48	13.50 ± 0.45	113.18	Sensitive	20.50 ± 0.77	18.65 ± 0.46	109.91
Chlorina	12.00 ± 0.28	11.65 ± 0.29	103.00	Insensitive	16.12 ± 0.36	16.12 ± 0.32	105.91

In wheat, out of 133 Norin varieties tested by Yamada (1991), 103 were GA-insensitive and 30 GA-responsive with all the 6 breeding lines being GA-insensitive. On

the other hand, out of the 16 landraces tested, 10 were GA-insensitive and 6 GA-responsive. One of the possible mechanisms of GA_3 action appears to be the promotion of changes in RNA synthesis during cell elongation. Apparently, GA_3 action in all target tissues may be via its effect on the transcriptional process. GA_3 may be enhancing the rate of synthesis of all classes of RNA, or it may be inducing some of the specific enzymes as in cereal aleurone.

The many effects of gibberellins suggest that they have more than one important primary site of action. Thus far, research with hormone receptors neither verifies nor denies that idea. Even a single effect such as enhanced stem elongation on whole plants results from at least three contributing events – (i) cell division is stimulated in the shoot apex, (ii) gibberellins sometime promote cell growth because they increase hydrolysis of starch, fructans, and sucrose into glucose and fructose molecules, and (iii) gibberellins sometime increase wall plasticity (Salisbury and Ross 2001).

The positive association between GA-insensitivity and height reduction has been shown in wheat for both *Gai*$_1$ with *Rht*$_1$ and *Gai*$_2$ with *Rht*$_2$ in crosses involving Norin 10 derivative semi-dwarfs (Gale and Marshall 1973, Hu 1974) and for *Gai*$_3$ with *Rht*$_3$ in 'Tom Thumb' type dwarf (Gale et al. 1975, Fick and Qualset 1975). Whether the nature of this association is pleiotropic, i.e. the *Gai* and *Rht* genes are, in fact, the same, or due to linked loci, as suggested by Konzak et al. (1973), it is clear that the chromosomal location of *Gai*$_1$, *Gai*$_2$,and *Gai*$_3$ are the same as those of *Rht*$_1$, *Rht*$_2$ and *Rht*$_3$. The exact nature of the relationship between gibberellin insensitivity and height reduction is of practical importance to wheat and barley breeders who are using different sources of dwarfism.

References

Fick, G.N. and C.O. Qualset, 1975: Genetic control of endosperm amylase activity and gibberellic acid responses in standard height and short strawed wheats. Proc. National Acad. Sci. U S A 72, 892-895.

Gale, M.D. and G. A. Marshall, 1973: Dwarf wheats and gibberellins. Pages 513-519 in Proc. 4th Wheat Genetics Symp.

Gale, M.D., C.N. Law, G.A. Marshall and A.J. Worland, 1975: The genetic control of gibberellic acid insensitivity and coleoptile length in a dwarf wheat. Heredity 34, 393-399.

Hu, M.I., 1974: Genetic analysis of semi-dwarfing and insensitivity to gibberellin GA_3, in hexaploid wheat. Ph D thesis, Washington State University, USA.

Konzak, C.F., Sadam Mir and E. Donaldson, 1973: Inheritance and linkage in durum wheats of semi-dwarfing genes with low response to gibberellin GA_3. Pages 29-40 in Proc. Symp. 'Genetics and Breeding of Durum Wheats' (Bari, 1973)

Morris, R., J.W. Schmidt and V.A. Johnson, 1972: Chromosomal location of a dwarfing gene in Tom Thumb wheat derivative by monosomic analysis. Crop Sci. 12, 247-249.

Salisbury, F.B. and C.W. Ross, 2001: Plant Physiology. CBS Publishers & Distributors, New Delhi (India).

Yamada, T., 1990: Classification of GA response, *Rht* genes and culm length in Japanese varieties and land races of wheat. Euphytica 50, 221-239.

Barley Genetics Newsletter 36:6-9 (2006)

Cytological investigation of winter two-rowed barley cv. Castor, the first Bulgarian cultivar of high cold resistance

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Absract
The cytological investigation comprises diakinesis, metaphase I, anaphase I and telophase II. In diakinesis 14.3% of the cells examined contain 6 closed bivalents and 1 open bivalent. The latter forms a rod bivalent in metaphase I. In anaphase there are laggard chromosomes in 5.8% of the cells while 7.0% of the cells contain anaphase bridges. There are not disturbances in telophase II and the meiotic stability is a prerequisite for high yield of about 800-850 kg/dca (a field trial).

Key words: barley, meiosis, open bivalent, anaphase bridge

Two-rowed barley cultivars occupying arable area in Bulgaria, belong to the winter-spring cereals which are highly productive and of low cold resistance. In severe winters with poor snow most barley plants die resulting in large economical loses. In 1993-2003 Prof. S. Tsvetkov working at the Dobrudja Agricultural Institute released the first Bulgarian two-rowed barley cultivar Castor possessing exclusively high cold resistance along with good productivity. The cytological analysis of the cultivar and the relationship between meiosis and grain yield were the subject of this study.

The standard aceto-carmine method was used to examine 35 cells for diakinesis, 145 cells for metaphase I, 170 cells for anaphase I and 200 tetrads for telophase II. Table 1 provides data indicating that in diakinesis over 85% of the cells contain 7 closed bivalents while 14.3% of the cells contain one open bivalent, associated with nucleolus. The phenomenon has been observed in other investigations (Stoinova, 1994; Stoinova and Tsvetkov, 2004).

The data concerning metaphase I indicate that almost 80% of the cells contain 7 closed bivalents/cell. The open bivalent in diakinesis forms a rod bivalent at metaphase I in 17.3% of the cells (Fig. 1a). Five closed and 2 rod bivalents were scored in four cells, while in one cell four closed and 3 rod bivalents were scored. Neither univalents nor multivalent associations were observed.

There were laggard chromosomes and bridges in anaphase cells (Fig. 1b and 1c). The laggards form genetically inactive micronuclei at telophase II only in single tetrads. At that phase cell partitions are properly situated, except for one preparation where 2 triads were observed. The presence of anaphase bridges was more interesting as their emergence is associated with paracentric inversion in heterozygous state. Synapsis between inverted and non-inverted chromosomes is accompanied by loop formation in pachytene. In the case of crossing over in that inversion loop anaphase bridges and fragments may be formed.

Anaphase bridges may result from gamma irradiation with 5 to 50 kR of barley chromosomes, the bridge frequency increasing with irradiation dose (Chandra and Makde, 1982).

The barley ring chromosomes may also lead to anaphase bridges in about 10% of the assayed cells (Singh and Tsuchiya, 1981). Bridge configurations have been found in tumor cells as well (Hoffelder, et al., 2004).

Disturbances at anaphase I in plant cells depend on environmental temperature. In two bean cultivars the disturbances have been found to vary from 34.5% to 58% at 22-26° C and decrease to 22-31% at 15-19° C (Konstantinov, 1971). Sakata et al. (2000) have shown that in barley the early stage until meiosis of PMCs is the most sensitive to high temperature stress. The stress may cause emergence of abnormal and irregularly shaped pollen grains at the heading stage. In cultivar Castor the disturbances at anaphase I are most probably due to high temperature during spike fixation, since chromosome reconstruction of inversion type proceed with very low frequency under normal environment, in the absence of chemical or physical mutagens and no ring chromosomes have been observed. The lack of disturbances at final meiosis phase is a prerequisite for normal pollen production and pollination as well as for high grain yield (up to 800-850 kg/dca a field trial).

The data from this cytological investigation suggest that meiosis in cultivar Castor proceeds without formation of univalents and multivalent associations at metaphase I with low percentage of laggard chromosomes and anaphase bridges while micronuclei may be present only in a single tetrads.

Table 1. Analysis of meiosis in barley Castor

Diakinesis				Metaphase I				Anaphase I					
Cells with 7closed bivalents		Cells with 6 closed + 1 open bivalent		Cells with 7 closed bivalents		Cells with 6 closed + 1 rod bivalent		Normal cells		Cells with laggard chromo-somes		Cells with bridge configur-ation	
n	%	n	%	n	%	n	%	n	%	n	%	n	%
30	85.7	5	14.3	119	82.7	26	17.3	148	87.2	10	5.8	12	7.0

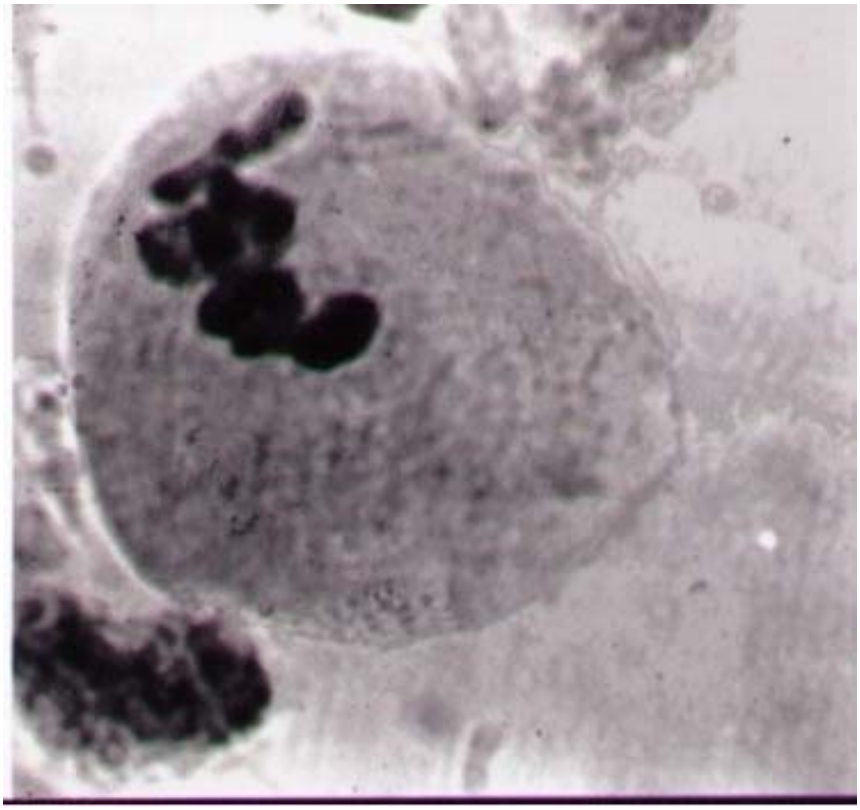

Fig. 1a. Metaphase I with rod bivalent

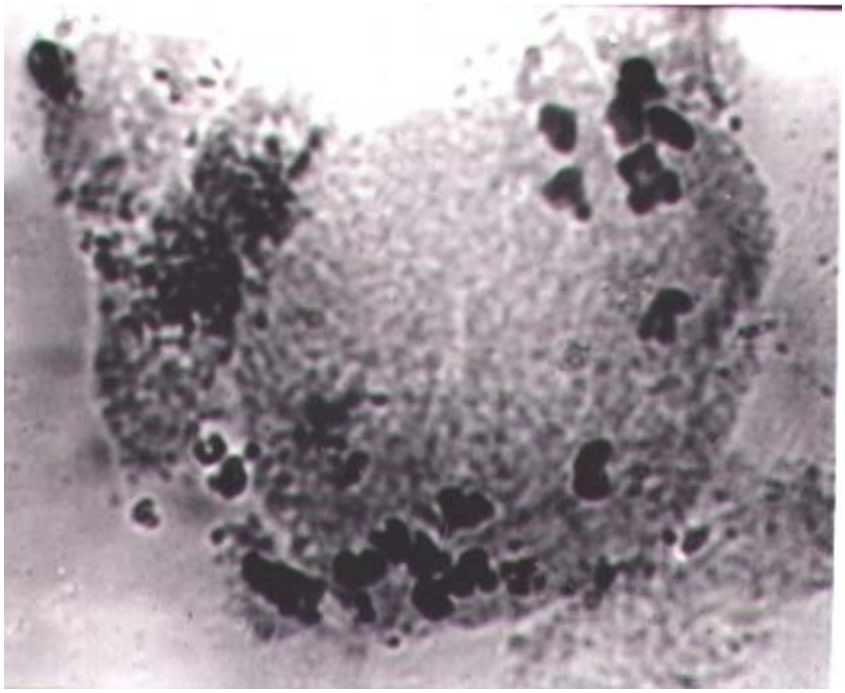

Fig.1b. Anaphase I with laggard chromosome

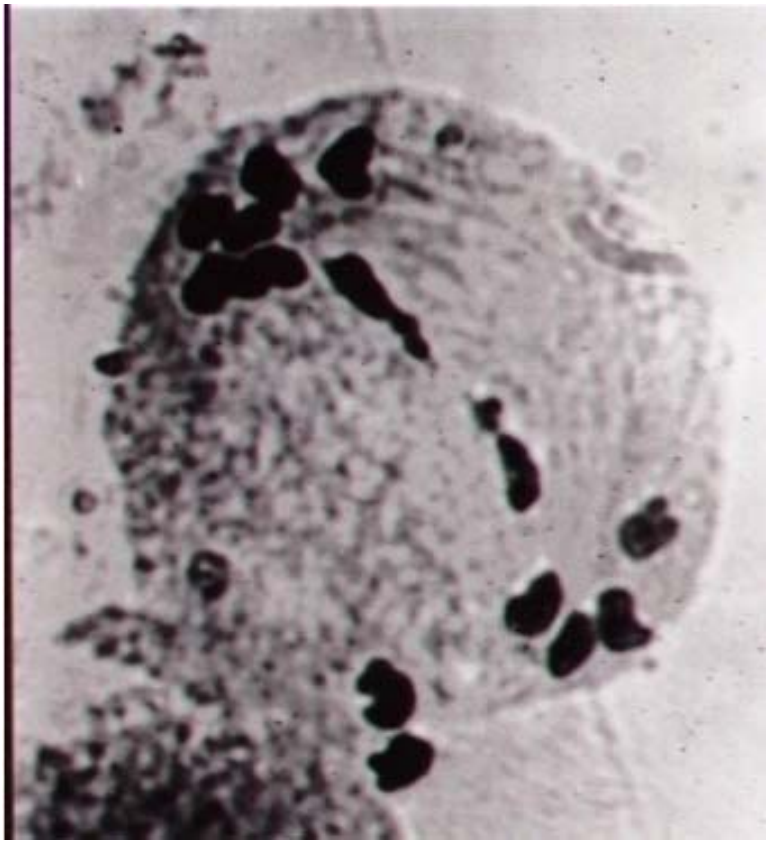

Fig.1c. Anaphase I with bridge configuration

References:

Chandra, A. and Makde, K. H. 1982. The rupture of radiation including bridges in barley. BGN 12: 11-13.

Hoffelder, D. R., Luo, L., Burke, N .A., Watkins, S. C., Gollin, S. M. and Sanders, W. S. 2004. Resolution of anaphase bridges in cancer cells. Chromosoma 112:389-397.

Konstantinov, A. V. 1971. Meiosis. Minsk, 18-19.

Sakata, T., Takahashi, H., Nishiyama, I. and Higashitani, A. 2000. Effects of high temperature on the development of pollen mother cells and microspores in barley, *Hordeum vulgare* L. J of Plant Res. 113:395-402.

Singh, R. J. and Tsuchiya. 1981. Ring chromosomes in barley (*Hordeum vulgare* L.). Chromosoma 82:133-141.

Stoinova, J. 1994. Characteristics of meiosis in some winter barley varieties. Cytologia 59:423-426.

Stoinova J. and Tsvetkov, S. 2004. Meiosis and productivity relationships in Bulgarian winter barley lines. BGN 34:5-9.

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Estimates of Heritability of Major Malting Quality Traits in Canadian Barley

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As part of the evaluation of potential new cultivars of barley in western Canada, 10 malting quality traits are assessed, at multiple sites in multiple years, on the most advanced malting lines in the co-operative testing system. These traits include thousand kernel and test weight, percent plump (over a 2.3 mm screen), percent crude protein, percent malt extract, percent soluble wort protein, diastatic power, level of alpha amylase and beta-glucans, and wort viscosity. Breeders focus on improvement of these traits, which are under quantitative genetic control and are the major characteristics in the improvement of malt quality (Bertholdsson, 2004).

The above malting traits are under both genetic control and environmental influence. For this reason, breeders estimate the likelihood and rate of genetic gain using (broad-sense) heritability (H^2; Strickberger, 1968). Although H^2 has been estimated for malting quality traits in a wide range of environments (Bertholdsson, 2004), there is little information on heritability of malting quality traits for (western) Canadian malting barley cultivars, including overlapping U.S. cultivars grown in the Northern Great Plains, and tested in western Canada.

Using data from the western Canadian co-operative barley trials from 2001 to 2005, a total of 120 genotypes were assessed for 10 malting quality traits in 32 environments. Broad-sense heritability (H^2) estimates were generated for each trait in each of the 5 years of evaluations (Table 1).

Table 1. Broad-sense heritability (H^2) for 10 malting barley quality traits for 120 genotypes in 32 environments under western Canadian conditions.

	Year							
Trait	2001	2002	2003	2004	2005	Avg.	Range	Std Error
1000 Kernel Weight	72.2	63.7	75.2	65.9	65.3	68.5	8.5	4.4
Test Weight	80.0	70.6	82.7	79.5	72.1	77.0	12.1	4.7
Percent Plump	52.7	75.2	49.9	79.5	61.6	63.8	29.6	11.8
Percent Crude Protein	79.1	53.4	94.0	78.1	82.7	77.5	33.8	13.3
Percent Malt Extract	77.9	87.2	53.2	67.9	75.8	72.4	40.0	11.4
Soluble Protein	72.6	77.1	34.5	79.4	71.8	67.1	44.9	16.5
Diastatic Power	72.1	76.8	42.9	58.0	84.1	66.8	41.2	14.7
Alpha-Amylase Levels	85.6	79.9	60.5	80.5	86.8	78.7	23.6	9.5
Beta-Glucans	64.9	63.7	51.5	60.6	72.8	62.7	21.3	6.9
Viscosity	78.8	49.5	69.8	82.1	63.7	68.8	32.6	11.6

Results in Table 1 show that the most stable traits, over years, are 1000 kernel weight, test weight, alpha-amylase and beta-glucans. Of these, test weight and alpha-amylase also have a high average heritability ($H^2 > 70$). Thus, these two traits are most likely to demonstrate a relatively rapid genetic gain from selection under western Canadian conditions. Crude protein

also demonstrated a high average heritability of 77.5, but was less stable, indicative of a fairly strong environmental influence. Thus, genetic gains would lag behind test weight and alpha amylase. The remaining seven traits had an average heritability value exceeding 62, indicating a positive response for genetic gain and are amenable to genetic improvement under western Canadian conditions, albeit at a slower pace. In addition, four out of the five years of testing were consistent, with respect to H^2, with 2003 being the exception. This indicates that genetic response may not be positive under a certain set of environments (year effect), underscoring the need to maximize multi-site, multi-year testing to realize improvement in malting quality.

References:

Bertholdsson, N.O. 2004. The use of environmentally stable grain characteristics for selection of high extract yield and low b-glucans in malting barley. Eur. J. Agron. 20(3): 237-245.

Strickberger, M.W. 1968. Genetics. MacMillan Publishing, N.Y. pp. 297-298.

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SSR Linkages to Eight Additional Morphological Marker Traits

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Introduction

More than 1000 morphological markers have been identified in barley (Franckowiak and Lundqvist 2002). These phenotypic traits were observed as spontaneous or artificially generated mutants in a wide range of cultivars over decades of research. Traits identified with alternate alleles include plant height, spike morphology, and seed size, among others. Approximately 275 of these markers have been placed on the morphological marker linkage map of the seven barley chromosomes (Franckowiak unpublished 2002) and/or on the consensus molecular marker linkage map (Kleinhofs 2002); approximately 150 to 200 additional markers have been placed on other maps but the rest have not been mapped.

Simple sequence repeat (SSR) markers are PCR-amplified regions of two or three base DNA repeats. Primers were designed to anneal to DNA on either side of each repeated segment, so size differences in amplification products are caused by different numbers of repeats in different genotypes. PCR reactions are easy to set up, do not use any hazardous chemicals, and only take a few hours for amplification. Products can be separated on agarose or acrylamide gels and results can be obtained in one day, which make these molecular markers ideal for short-term projects. We have worked with high school students in the North Dakota Governor's School to map morphological traits over the last several summers. This report describes the location of eight additional morphological markers through linkage to SSR markers.

Materials and Methods

Mapping population development A set of mutant lines were selected from various barley collections based on phenotype and backcrossed four to six times to 'Bowman' (Table 1). Homozygous backcrossed lines, BC_nF_2 populations, and BC_nF_2-derived F_3 lines were developed for mapping with molecular markers. Fifty BC_nF_2 seeds were sown for each mutant line in a greenhouse, along with the homozygous mutant parent and Bowman. The greenhouse was maintained at 21-26^oC with a 16 h day/8 h night cycle supplied by sodium halide lights. Each plant was scored as either normal or mutant. Plants were grown to maturity and harvested. Twelve BC_nF_3 seed were sown for each BC_nF_2 plant to identify heterozygous lines. Parent and BC_nF_2 seed were sown one per 15 cm clay pot, in a soil-less potting mix supplemented with a slow release fertilizer (14-14-14). The BC_nF_3 seed were sown with six seed per 15 cm clay pot and scored for the mutant trait at the appropriate time. Plants were treated with Marathon (Imidaclorprid) systemic insecticide at approximately the 2-3 leaf stage.

Trait Analysis Backcross-derived near-isogenic lines (NILs) and Bowman were planted in a field near Christchurch (Leeston), New Zealand and near Aberdeen, ID in 2002 and 2003 for agronomic analysis. Height (cm) and lodging (1-9 scale) were measured at Christchurch only in both years at the hard dough stage. Peduncle (cm) and awn length (cm), and the number of kernels per spike were measured in all four environments. Leaf length (cm) and width (mm) were measured on the penultimate leaf blade in all environments except 2002 Christchurch.

Rachis internode length (mm) was measured in all environments except 2003 Aberdeen. Kernel weight (mg per kernel based on 100 kernels) was measured in all environments except 2002 Aberdeen. Yield (g/m) was measured at Christchurch in 2002 and Aberdeen in 2003. Heading date was measured at Aberdeen in 2002. Lines also were grown in Dundee, Scotland in 2005, measuring heading date, height, peduncle and awn length, rachis internode length and number of kernels per spike. Plots were sown in an augmented block design with Bowman repeated across blocks. Each plot was a 2-m row with rows spaced 60 cm apart. The seedling rate was approximately 20 seeds per meter of row. Data were analyzed by Analysis of Variance using SAS (SAS Institute Inc. 2002) and least squares means were compared using the General Linear Model procedure. Trait means of the near-isogenic lines were compared to Bowman means using Least Significant Difference values.

Molecular marker testing Leaf tissue was harvested from young parent and BC_nF_2 plants grown in the greenhouse and the DNA extracted using the method of Dahleen et al. (2003). The DNA was then resuspended in 200 µL of modified TE (10 mM Tris-Cl, pH 7.4 and 0.1 mM EDTA). Simple sequence repeat (SSR) markers (Ramsay et al. 2000) were screened against the mutant near-isogenic lines and Bowman to identify polymorphisms. The PCR methods used were described in Dahleen et al. (2003). Amplified fragments were separated by gel electrophoresis using 4% Super Fine Resolution (SFR) agarose (Amresco, Solon, OH) in1 X TAE (40 mM Tris-acetate and 1 mM EDTA). Alternatively, acrylamide gel electrophoresis was used as described by Wang et al. (2003). The gels were stained with ethidium bromide, and photographed under UV light. Markers that detected polymorphisms between Bowman and a mutant near isogenic line were tested on the corresponding BC_nF_2 population. SSR and mutant trait segregation data were entered into MAPMAKER software (Lander et al. 1987; Lincoln et al. 1992) to test linkage between the markers and the mutant trait.

Table 1. Morphological markers, Barley Genetic Stock number, their chromosome locations, pedigrees of the mapping populations and the number of backcrosses to Bowman to develop the near-isogenic lines

Gene	BGS No.	Chromosome	Pedigree	Backcrosses
cer-zt.389	BGS437	2H	cer-zt.389/5*Bowman	4
dsp.ah		7H	DWS1180 Mut4841/6*Bowman	5
dsp.at		3H	Bowman*5/DWS1220 7117	4
dsp.ba		7H	DWS1357 UT1713/6*Bowman	5
int-k.47	BGS546	7H	int-k.47/7*Bowman	6
nec.50 (*pmr2*)	BGS634	7H	Bowman*5/nec.50	4
nec.54 (*pmr2*)	BGS634	7H	Bowman*5/nec.54	4
pyr.ai		3H	Bowman*6/DWS1018	5

Results and Discussion

Linkage between SSR markers and the morphological traits was identified for each of the eight backcrossed morphological marker lines. All F_2 populations segregated 3:1 normal:mutant as expected for single recessive traits. There were no significant differences between Bowman and near-isogenic lines for lodging, leaf width or heading date.

The eceriferum mutant *cer-zt*.389 (BGS 437) was located approximately in bin 1 of chromosome 2H, linked to Bmac0134 (Fig. 1). This NIL only differed from Bowman for kernels per spike, with a significant increase in *cer-zt*. The intermedium spike mutant *int-k*.47 (BGS 546) was located in the centromeric region of chromosome 7H, closely linked to markers Bmag0217 and Bmac0162 in bins 6 to 7. The *int-k*.47 NIL had significantly reduced height, peduncle length, awn length, kernels/spike, leaf length, kernel weight, and yield. The two necroticans mutants (*nec.50* and *nec.54*) are alleles at the premature ripe 2 (*pmr2*) locus (BGS 634). Analysis of individual populations indicated both were linked to the same markers on chromosome 7H (Fig. 1). Because they are alleles, a combined linkage analysis was conducted, which showed tight linkage to markers in bin 5 of 7H (Fig. 2). The *nec.54* NIL was analyzed in the field and showed reduced yield compared to Bowman. The pyramidatum mutant *pyr.ai* was linked between markers on chromosome 3H in bins 5 to 7 (Fig. 1). The NIL was shorter than Bowman, had shorter rachis internodes, and showed a significant increase in kernels per spike.

Three additional dense spike mutants were mapped, *dsp.ah*, *dsp.at*, and *dsp.ba*. The first two were linked to markers on chromosome 7H near the centromere while *dsp.ba* was located near the centromere on the short arm of chromosome 3H (Fig. 1). Previously located dense spike loci include *dsp1* on chromosome 7HS, *dsp9* on chromosome 6HL, and *dsp10* on chromosome 3HS. Agronomic trait performance of these three lines was similar to Bowman for most traits. The *dsp.ah* mutant showed significantly reduced height, peduncle length and rachis internode length, and lower yields but showed a significant increase in the number of kernels per spike. The *dsp.at* mutant was shorter and had reduced peduncle and rachis internode lengths, while *dsp.ba* only showed a decrease in rachis internode length compared to Bowman. These differences in the *dsp* mutants indicate it is unlikely they are alleles, and they are unlikely to be alleles of *dsp1*, *ert-a, ert-d* or *ert-m*, mutants with similar dense spikes.

In summary, we have placed eight additional morphological mutants on the barley molecular linkage map. One (*cer-zt*) mapped to chromosome 2H, two (*dsp.ba* and *pyr.ai*) mapped to chromosome 3H, and five (*dsp.ah*, *dsp.at*, *int-k*.47, *nec*.50 and *nec*.54) mapped to chromosome 7H.

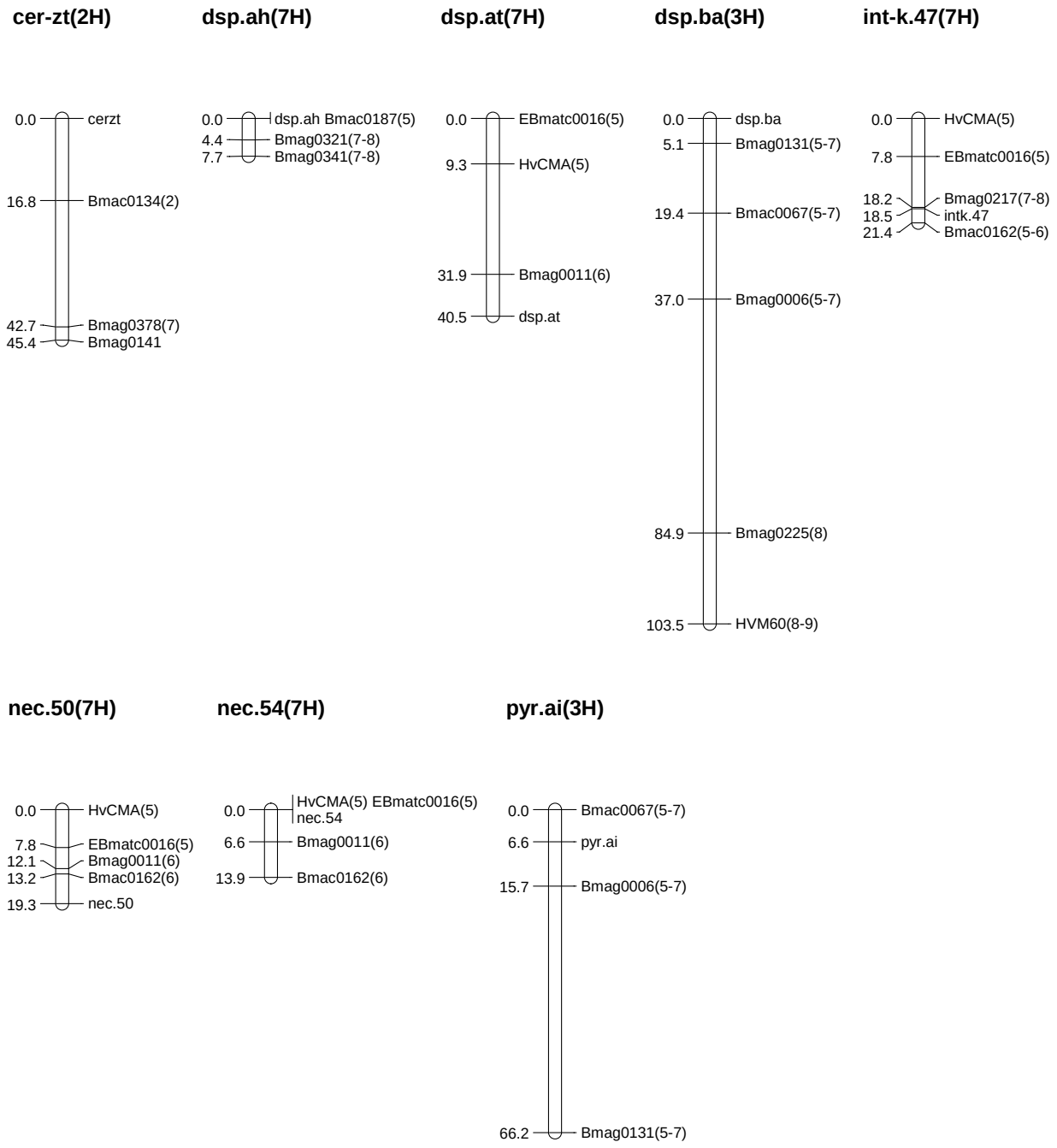

Figure 1. Linkage maps showing positions of morphological markers in relation to SSR markers mapped in segregating populations of 50 F_2 plants for each trait. Cumulative linkage distances are on the left of the vertical bar and marker names are on the right. Approximate chromosomal bin locations for the SSRs are in parentheses.

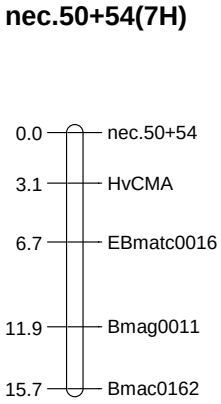

Figure 2. Linkage map showing the position of the *nec*.50 and *nec*.54 when the trait and marker data were combined for these alleles at the *pmr2* locus (BGS 634).

References

Dahleen LS, Franckowiak JD and Vander Wal LJ. 2003. Exposing students and teachers to plant molecular genetics with short-term barley gene mapping projects. J Natural Resour Life Sci Educ 32:61-64.

Franckowiak JD and Lundqvist U. 2002. Current list of new and revised barley genetic stock descriptions [Online]. Available at http://wheat.pw.usda.gov/ggpages/bgn/32/index.html (verified 18 May 2006). Barley Genet Newsl 32:47–137.

Kleinhofs A. 2002. Integrating molecular and morphological/physiological marker maps [Online]. Available at http://wheat.pw.usda.gov/ggpages/bgn/32/index.html (verified 18 May 2006). Barley Genet. Newsl. 32:152–159.

Lander E, Green P, Abrahamson J, Barlow A, Daley M, Lincoln S, and Newburg L. 1987. MAPMAKER: an interactive computer package for constructing primary genetic linkage maps of experimental and natural populations. Genomics 1:174-181.

Lincoln S, Daly M and Lander E. 1992. Constructing genetic maps with MAPMAKER/EXP 3.0. Whitehead Inst. Tech. Rep. 3rd ed. Whitehead Inst., Cambridge, MA.

Ramsay L, Macaulay M, Ivanissevich S, MacLean K, Cardle L, Fuller J, Edwards KJ. Tuvesson S, Morgante M, Massari A, Maestri E, Marmiroli N, Sjakste T, Ganal M, Powell W, and Waugh R. 2000. A simple sequence repeat-based linkage map of barley. Genetics 156:1997-2005.

Wang D, Shi J, Carlson SR, Cregan PB, Ward RW, and Diers BW. 2003. A low-cost, high-throughput polyacrylamide gel electrophoresis system for genotyping with microsatellite DNA markers. Crop Sci 43:1828-1832.

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Expression of a P450 gene in barley (*Hordeum vulgare*)

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The sequence reported here was submitted to GenBank (Accession number DQ838490)

Abstract

A new member of the P450 gene family was identified in barley. The P450 gene was a member of the subfamily CYP72A. Northern analysis revealed that various CYP72A39 transcripts were found in four barley cultivars at a very early vegetative stage but no expression was detected at the reproduction stage. This suggested that this P450 gene may be involved in seedling development in barley. Comparison of expression profiles of this gene and "digital expression" databases confirmed that this gene was homologous to several cereal EST clones with tissue-specific transcripts responding to various environmental stimuli. Among these, many transcripts in barley were obtained from stressed tissues at the vegetative stage, and two transcripts in wheat were expressed after being challenged by barley powdery mildew pathogen (*Bumeria graminis* f. sp. hordei). This suggested that the CYP72A39 gene may play a role in defence in the barley seedling.

Introduction

Plant P450s belong to the cytochrome group of membrane-bound enzymes, that are usually found in plant endoplasmic reticulum (Halkier, 1996). Among 6008 P450 genes reported in all organisms including human, one third of them have been reported in plant species. In plants, P450s play a central role in numerous biosynthetic pathways such as production of second metabolites (phenylpropanoids, alkaloids, lipids, sterols, flavonoids and cyanogenic glycosides). In addition, plant P450s have a significant response to stresses (Whitbred and Schuler, 2000) and disease (Smigocki and Wilson, 2004; Takemoto et al., 1999). P450 enzymes that are encoded by P450 genes are important components of the defense mechanism of plants through their enzymatic reactions in various biosynthesis pathways. Depending on the type of stimuli, specific enzymes are produced under stress conditions. Many reports (Bolwell et al., 1994; Halkier, 1996; Mizutani et al., 1998; Persans et al., 2001; Schuler and Wreck-Reichhart, 2003; Toguri et al., 1993c) have emphasized the affects of environmental factors such as stresses (salinity, low temperature, pathogen elicitation) as well as developmental factors (plant age, tissue specific) on the expression pattern of plant P450. Here we report the identification and analysis of expression patterns of a new P450 gene in barley.

Materials and methods

Randomly selected clones (512) from a cDNA library of four day old seedling (cv Alexis) were sequenced. Details of the cDNA library construction were previously described by Holton and co-workers (2002). A clone (5E2) was identified showing 72% homology to a P450 (CYP72A21) in rice.

This 1817bp full length sequence, consisting of 1578 nucleotide ORF, 189 nucleotides at the 3' UTR and 50 nucleotides in the promoter region, encoded a 526 amino acid polypeptide with molecular mass of 59.57 kDa and isoelectric point of 8.59. Coding sequence was searched against the P450 database from David Nelson's home page (http://drnelson.utmem.edu/CytochromeP450.html) using BLASTP. P450 motifs and domains in the sequence were detected manually.

Four parental barley lines (Chebec, Harrington, Alexis and Sloop) which have been used in many breeding programs were selected. Two growth conditions, indoor and outdoor, were applied to each cultivar. Observation and sample collection were based on the Zadok score, a measurement of plant growth stage. For each cultivar, samples were collected at different development stages, ranging from young seedling to post-anthesis (Table 1). For all four cultivars, at vegetative stage, both roots and shoots were collected for 4 day and 1 week old seedling samples, and only shoot samples for 2 week old seedling (Parts A, B, C of Figures 1 and 2). At the floral stage, only inflorescence parts were sampled for two cultivars, Chebec and Harrington. Flowering was checked daily and the first appearance of the flag leaf, indicating that the plant was already in its floral stage, was marked. The floral stage (Parts C, D of Figures 1 and 2) was divided into four sub-stages: stage 1 was when the flag leaf was first seen, stage two was when the first awn appeared and stage three was recorded if the first spikelet was seen. These three stages occurred before plants pollinated. When pollination occurred, individual seeds that were pollinated on that day were marked. Two weeks later, these marked seeds were harvested and classified as at sub-stage four.

Total RNA was extracted from the tissues indicated in Table 1 using either RNeasy Plant Mini kit (Qiagen) or CTAB method (Chang et al., 1993). Probes were generated by using DIG-PCR probe synthesis kit (Roche). Membrane transfer, blot hybridisation and detection were carried out according to the manufacturer's protocol (Roche, 2000). Barley Ubiquitin was used as house keeping gene. RNA extracted from mix-tissues of shoot and root (4 day seedling) was used as positive control. Primer sequences and PCR compositions to generate labelled probes were listed in Tables 2 and 3.

The temperature cycling condition for both probes were very similar. They were 96^0C for 2 min; followed by 25 cycles of 96^0C for 1min, 45^0C for 45s, 72^0C for 2min, and a final extension at 72^0C for 10min. The only exception was that the duration of extension at 72^0C was 1min shorter for the whole 25 cycles when the Ubiquitin primer was used.

Results and discussion

Sequence structure of CYP72A39

Results of a BLAST search of CYP72A39 sequence against the Nelson's P450 database (http://132.192.64.52/p450.html) showed 72% and 68% identity to CYP72A21 and CYP72A33 in rice respectively. This indicated that this new barley gene belong to the CYP72A subfamily and was assigned as CYP72A39.

Several highly conserved regions which are consider as a benchmark of P450s (Fig 3) are found in the gene structure of the new sequence such as:

- The membrane anchor (MVLLGVLASPTPATVLWTLLGLALL) is situated in the first 25 amino acids from the N terminal,

- The C-helix motif (WVKHR) is located from residue 157 to161,
- The I-helix motif (AGSET) required for oxygen binding, is located from residue 335 to 339,
- The K-helix motif (EVLR) is located from residue 391 to 394,
- The P450 signature PERF (also called domain C) is located from residue 446 to 449,
- The heme binding region PFGWGPRICIG (also called domain D) is located from residue 465 to 475 including the Cystein residue in position 473.

However, the proline rich motif (PPGP) which is usually located between the C-helix and the membrane anchor, was not detectable in the barley CYP72A39 sequence. Similarity, the absence of a proline rich motif in the protein structure of CYP72A1 (accession L10081) was reported in Catharanthus roseus (Irmler et al., 2000).

Gene expression patterns of CYP72A39 by Northern blot analysis

At the vegetative phase, under the same indoor condition; the CYP72A39 gene was expressed differently in all four cultivars. Overall, the CYP72A39 transcript was found in root of 1 week old seedling of all four cultivars (Figures 4, 5, 6). In cultivar Chebec, the gene was expressed only in the root of 1 week old seedling (Figure 4). No transcript was found in shoot at any stage of the vegetative phase of this cultivar. In contrast, in cultivar Alexis, bands were found only in root at 4 day and in 1 week old seedlings (Figure 6). Interestingly, the transcript of the candidate gene hybridised strongly to RNA extracted from mixed tissues (root and shoot at 4 day old seedling) of Alexis that were used as positive probe control in all blots, but there was no evidence of this candidate gene's transcriptions found in any shoot tissue of this cultivar. Furthermore, as shown in Figures 5 and 6, CYP72A39 was expressed in the shoots and roots of both Sloop and Harrington. At the floral stage, no hybridised bands were detected in any reproductive tissues of any cultivar, under both growth conditions, indoor and outdoor (Figures 4, 5, 6). The house keeping gene, Ubiquitin, was expressed in all tissues of the four cultivars at all developmental stages as expected.

Comparative analysis of gene expression of CYP72A39 and sequences from "electronic expression" databases

The gene expression patterns of CYP72A39 were compared to "electronic expression" data from plant species by homology search. BLAST results indicated that the CYP72A39 matched to numerous ESTs derived from several cDNA libraries (Table 4). However, only a fraction of entries (11 out of 171) are listed here.

Results of the Northern blot experiment showed that transcripts of CYP72A39 were specifically found only in the vegetative tissues of the four cultivars. This study also demonstrated that, in the two cultivars Chebec and Harrington, there was no evidence of CYP72A39 transcripts in any reproductive tissues, either from the growth cabinet or field grown barley. Further more, the blast search against Plant Genome database (Plant GDB) showed that CYP72A39 was highly homolog to EST sequences isolated at seedling stages. These factors suggest that this gene may play a role in seedling development of barley.

In most cases, the CYP72A39 showed significant homology to sequences whose transcripts were induced in vegetative tissues. Blast searches against the Plant GDB server showed several hits to the monocot EST sequences that appeared under stressed or pathogenic challenging. To get more specific sequence homology to each species, blast searches against individual EST database was performed. Results from the searches against the barley EST database showed that the

CYP72A39 had 100% identity (over 812/812 nucleotides) at both coding and the 3-UTR region with the barley contig20974 obtained from cold stressed shoots. In the coding region, the three motifs, K helix, C domain and D domain containing the Heme region that essential to a P450 gene, were identical to those found in the CYP72A39 sequence.

Results from the searches against the wheat EST database indicated that the CYP72A39 was strongly homologised to five wheat ESTs from the same cultivar, Fidel. These five wheat ESTs were listed as pathogenic inducible genes. Of these, two EST sequences AJ888598 and AJ890237 showed significant identity (93% over 594/638 and 627/674 nucleotides respectively) to the CYP72A39. Sequence comparison between these two and the CYP72A39 showed that they shared a 168 identical amino acid sequence included the stop codon (ATG). Similar to the barley contig20974 sequence, these two wheat ESTs also contained the three motifs (K helix, C and D domains) identical to those found in the CYP72A39 sequence (Figure7). Ten amino acid substitutions were found between CYP72A39 and the two wheat ESTs. Attention was focused on the AJ888598 sequence, the 3'UTR of which matched to that of the CYP72A39 sequence (data not shown). However at present, information about the wheat EST (AJ888598) was very limited. Overall significant sequence identity at both coding, particularly at the three important motifs, and non-coding regions between the CYP72A39 and several monocot ESTs suggested that the CYP72A39 gene appears to be a good candidate for stress and disease resistant study.

Conclusion

Various expression patterns of CYP72A39 genes were found in four barley cultivars by Northern analysis. These patterns detected in vegetative but not in any reproductive tissues suggested that this gene may be involved in seedling development of barley. In addition, significant homology between this gene and cereal EST samples expressed under stresses and particularly pathogen challenges leads to a speculation that functionally this barley gene is likely to be involved in defence in barley.

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References

Bolwell, G.P., K. Bozak, and A. Zimmerlin. 1994. Plant cytochrome P450. *Phytochemistry*. 37:1491-1506.

Chang, S., J. Puryear, and J. Cairney. 1993. A simple and efficient method for isolating RNA from pine trees. *Plant Mol Biol Reporter*. 11:113-116.

Halkier, B.A. 1996. Catalytic reactivities and structure/function relationships of cytochrome P450 enzymes. *Phytochemistry*. 43:1-21.

Holton, T.A., J.T. Christopher, L. McClure, N. Harker, and R.J. Henry. 2002. Identification and mapping of polymorphic SSR markers from expressed gene sequence of barley and wheat. *Molecular Breeding*. 9:63-71.

Irmler, S., G. Schroder, B. St-Pierre, N.P. Crouch, M. Hotze, J. Schmidt, D. Strack, U. Matern, and J. Schroder. 2000. Indole alkaloid biosynthesis in Catharanthus roseus: new enzyme activities and identification of cytochrome P450 CYP72A1 as secologanin synthase. *Plant Journal*. 24:797-804.

Mizutani, M., E. Ward, and D. Ohta. 1998. Cytochrome P450 superfamily in Arabidopsis thaliana - isolation of cDNAs, differential expression, and RFLP mapping of multiple cytochromes P450. *Plant Molecular Biology*. 37:39-52.

Persans, M.W., J. Wang, and M.A. Schuler. 2001. Characterization of maize cytochrome P450 monooxygenases induced in response to safeners and bacterial pathogens. *Plant Physiol*. 125:1126-1138.

Roche. 2000. DIG application manual for filter hybridisation. Roche Diagnostics GmbH, Mannheim/Germany. 225 pp.

Schuler, M.A., and D. Wreck-Reichhart. 2003. Functional genomics of P450s. *Ann. Rev. Plant Biology*. 54:629-657.

Smigocki, C.A., and D. Wilson. 2004. Pest and disease resistance enhanced by heterologous suppression of a Nicotiana plumbaginifolia cytochrome P450 gene. *Biotechnology Letters*. 26:1809 - 1814.

Takemoto, D., M. Hayashi, N. Doke, M. Nishimura, and K. Kawakita. 1999. Molecular cloning of a defense-response-related cytochrome P450 gene from tobacco. *Plant Cell Physiol.* 40:1232-1242.

Toguri, T., N. Umemoto, O. Kobayashi, and T. Ohtani. 1993c. Activation of anthocyanin synthesis genes by white light in eggplant hypocotyl tissues, and identification of an inducible P-450 cDNA. *Plant Mol Biol*. 23:933-946.

Whitbred, J.M., and M.A. Schuler. 2000. Molecular characterization of CYP73A9 and CYP82A1 P450 genes involved in plant defense in pea. *Plant Physiol*. 124:47-58.

Table 1. Developmental stages, type of collected tissues and growth conditions of barley materials used in the gene expression experiment.

Developmental stage	Morphological appearance	Type of tissue collected	Growth condition[a]	Cultivar[b]
Vegetative stage	Four day old seedling	Shoot, Root	I	C, H, A, S
	One week old seedling	Shoot, Root	I	C, H, A, S
	Two week old seedling	Shoot	I	C, H, A, S
Floral stage	Appearance of flag leaf	Head	I/O	C, H
	Appearance of first awn	Head	I/O	C, H
	Appearance of first spikelet	Head	I/O	C, H
	Two weeks after pollination	Individual seed	I/O	C, H

[a]I: Indoor; O: Out door;
[b]C: Chebec, H: Harrington A: Alexis, S: Sloop

Table 2. Primer sequences

Primer ID	Primer sequence (5'-3')	Probe length (bp)
CYP72A39 forward	TCAAGCACCGGAGGATCCTCA	1023
CYP72A39 reverse	CAAGGTCGAATTCGAAGCGTTGA	
Ubiquitin forward	CGACAACGTCAAGGCGAAGAT	314
Ubiquitin reverse	CCAAAGCCACGGCACAAGTT	

Table 3. PCR components of labelled and unlabelled samples

	For CYP72A39 probe		For house keeping gene probe	
	Labelled fragment	Unlabelled fragment	Labelled fragment	Unlabelled fragment
10x PCR buffer (1.5mM MgCl2, Roche)	2µl	2µl	2µl	2µl
Taq Polymerase (5 units, Roche)	0.4µl	0.4µl	0.4µl	0.4µl
10mM dNTP's (Promega)	_	0.4µl	_	0.4µl
DMSO	1µl	1µl	_	_
50% glycerol	1.2µl	1.2µl	_	_
PCR DIG mix (10x concentration, Roche)	2µl	_	2µl	_
Primer forward (10µM)	3µl	3µl	3µl	3µl
Primer reverse (10µM)	3µl	3µl	3µl	3µl
Plasmid DNA	10ng	10ng	100pg	100pg
And MiliQ water was added to a total volume	20µl	20µl	20µl	20µl

Table 4. Representative sequences that homolog to CYP72A39

Accession	Species	Cultivar	Expressed tissue	Description of treatment and sample source (if known)	Nucleotide identity %	E value
Contig 20974*	Barley	Morex	5 day old shoot seedling	Cold stress (EST and Chip)	100 (812/812)	00.0
BF623161	Barley	Morex	5 day old shoot seedling	Cold stress (EST and Chip)	98 (581/591)	0.00
AJ888598	Wheat	Fidel	Leaf of 7 day old seedling	Pathogenic elicitor (EST)	93 (594/638)	00.0
AJ890237	Wheat	Fidel	Leaf of 7 day old seedling	Pathogenic elicitor (EST)	93 (627/674)	00.0
CB871128	Barley	Sloop	3 day old coleoptile	Unknown (EST)	97 (412/423)	00.0
CB867923	Barley	Sloop	3 day old coleoptile	Unknown (EST)	97 (420/430)	00.0
BE587940	Rye	Blanco	Root tip of seedling	Unknown EST	92 (432/469)	e-180
BU050675	Maize	NA	NA	Unknown (EST, Unigen)	85 (372/433)	e-101
CN137181	Sorghum	BTx623	Root and leaf of 8 day old seedling	Methyl viologen treatment, oxidative stressed leaves & roots (EST)	84 (393/467)	2e-90
BQ743931	Wheat	Chinese Spring	Root at tillering stage	Salt stress (EST)	81 (414/508)	4e-67
CB927177	Sorghum	IS3620C	Seedling	Abscisic acid (ABA) treatment (EST)	84 (307/364)	1e-69

Asterisk (*) indicated blast results obtained from Barley1contig20974 of 3 members (HVSMEa0012A09f2, HVSMEa0019H07r2, HVSMEa0012A09r2). Unknown means not enough specific information to determine if tissues were treated or not.

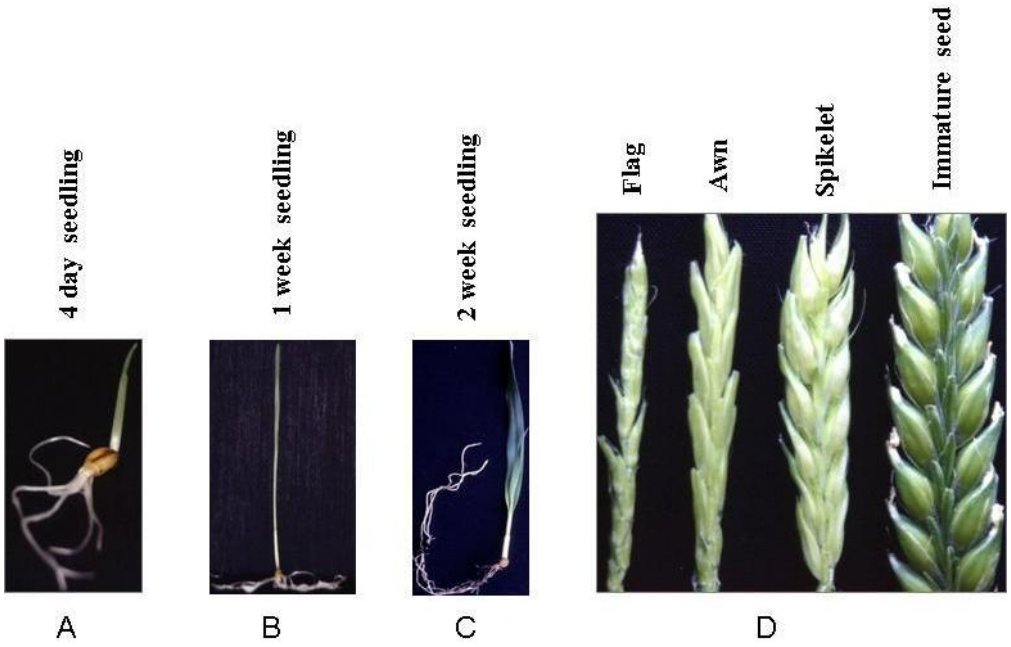

Figure 1. Development and appearance of cultivar Chebec at various sample collection stages. A, B, and C: seedlings at vegetative stage. D: inflorescence parts at floral stage of indoor plants.

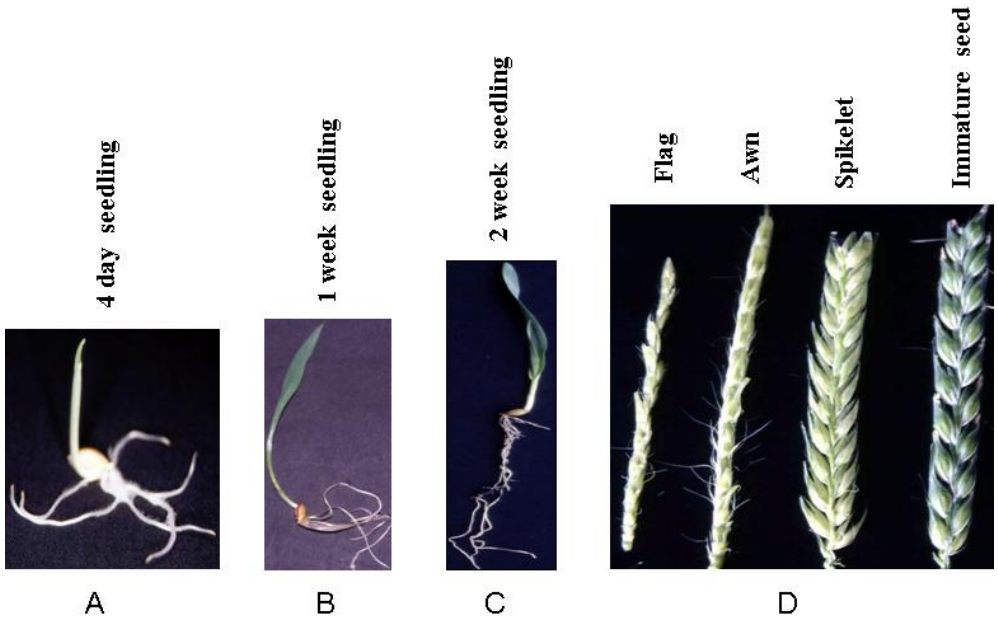

Figure 2. Development and appearance of cultivar Harrington at various sample collection stages. A, B, and C: seedlings at vegetative stage. D: inflorescence parts at floral stage of indoor plants.

```
MVLLGVLASPTPATVLWTLLGLALLWQVKRLVDYTWWRPRRLQRALRAQG -50
Membrane anchor

LRWFGGTPYRFPVGDLGDYGRQGKEASSRALPLRCHDIRAHVAPYLYSTV -100

LEHGKTCVSPVPKVTIADPGVTREVMSNKFGHFEKLQFPTLTRLLAGGVA -150

VYEGEKWVKHRRILNPAFHLEKLKLMMPAFSACCEELVSRWTQSLGSDGW -200
      C helix

CEVDVCPEFQTLTGDVISRTAFGSSYLEGRRIFELQSVQADRIVAEVKKI -250

FIPGYMSLPTKKNKLMHETNNEVESILRGLIEKRMQAMQQGETTKDDLLG -300

LMLESNMKETDDKGQPILGMTIEEVIEECKLFYFAGSETTSVLLTWTMIV -350
                                   I helix

LAMHPEWQDRAREEVLGLFGKNKPEYDGFSKLKTVTMILYEVLRLYPPAI -400
                                              K helix

AFMRKTYKEIEIGSITYPAGVIIELPVLLIHHDPDIWGSDVHEFKPERFA -450
                                                 Domain C

NGIAKASKDPGAFLPFGWGPRICIGQNFALLEAKMALCMILQRFEFDLAS -500
              Heme binding region

TYSHVPHNQKMLRPMHGAQIKLRAI* -526
```

Figure 3. Deduced amino acid sequence of CYP72A39 is shown in capital letters. Motifs and domains are underlined. Stop codon is indicated by asterisk (*).

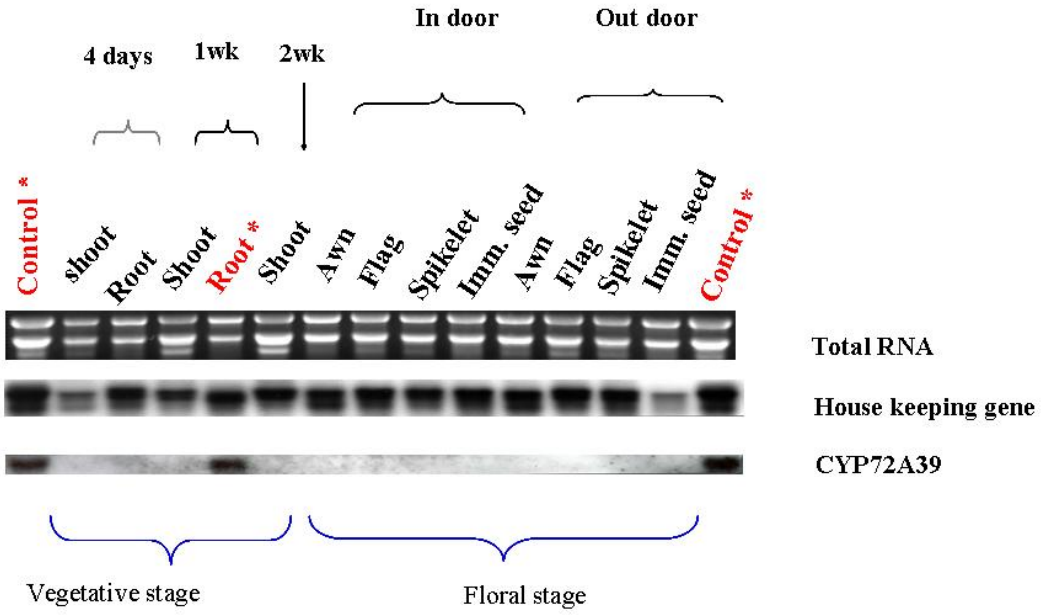

Figure 4. Expression patterns of CYP72A39 in cultivar Chebec by Northern blot analysis. Asterisk (*) indicates expressed tissues.

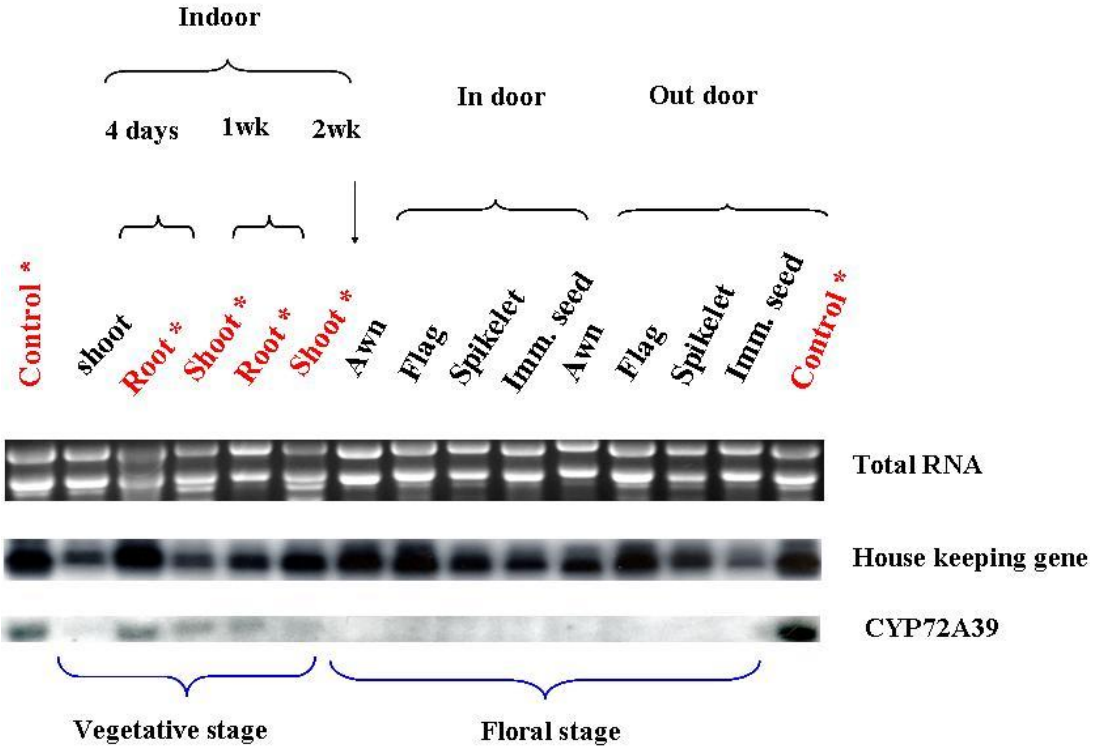

Figure 5. Expression patterns of CYP72A39 in cultivar Harrington by Northern blot analysis. Asterisk (*) indicates expressed tissues.

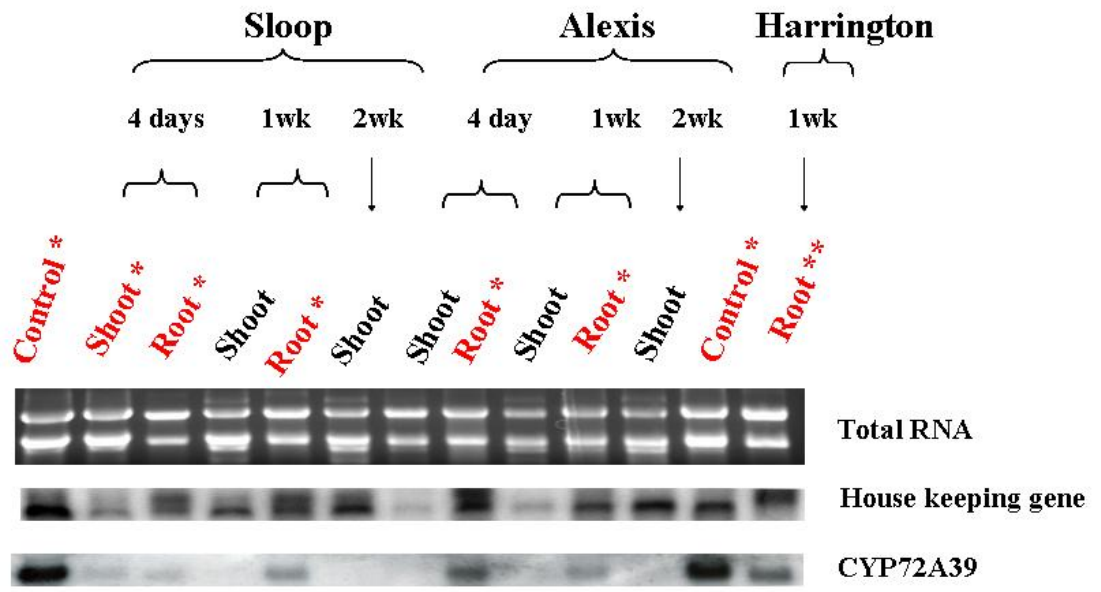

Figure 6. Gene expression patterns of CYP72A39 in cultivars Sloop and Alexis by Northern blot analysis. Asterisk (*) indicates expressed tissues. Double asterisk (**) indicates expressed tissue of 1 week old seedling root of Harrington (as shown in Figure 4) used as an extra internal control.

Figure 7. Alignment of CYP72A39 and the three EST sequences (barley contig 20974, wheat AJ888598 and AJ890237). Motifs are indicated by arrows. Mismatched sequences are indicated in colon (:). Stop codon is indicated by asterisk (*).

Barley Genetics Newsletter 36:28-29 (2006)

Description and technological characteristic of the new winter two-rowed barley cultivar Caskadior 3

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Professor S. Tsvetkov and team, working at the Dobruja Agricultural Institute released cultivar Caskadior 3. The new winter barley cultivar formed compact crops with good stability to lodging, grain falling out and spike brittleness. It is presented in Fig.1. Caskadior 3 is usually sowed in autumn, but under extremely conditions the plants are successfully grown and in early spring sowing.

The plants were inoculated with spore suspension of leaf and stem rusts and powdery mildew and showed high resistance to those diseases. The cultivar possesses also high drought resistance. During 2001 under the conditions of extremely high soil dryness in the area of Dobrudja Agricultural Institute is yielded 550 kg/dca average versus 240 kg/dca for wheat. Because of these traits Caskadior 3 is suitable for growing in the whole country.

The data from the State Variety Testing manifest that Caskadior 3 produced higher yield than the standard cultivar Obzor – 14.8%(95 kg/dca field trial). Its productive yield potentiality in normal years is over 850 kg/dca (the field trial). The precondition for the high yield obtained is also the meiotic stability of cultivar Caskadior3. In telophase II only 1.33% of the analysed over 300 tetrads had micronuclei. On the other hand Maich and Ordonez (2003) considered that systematical plant breeding program directed to increase grain yield simultaneously improved the meiotic performance of hexaploid triticale.

Caskadior 3 formed very large uniform (96.4%) kernels with 1000 kernel weight of 55 grams. Because of that it may be possible with only one cleaning of the raw material to be given to the breweries. The protein content is 9.7% versus 10.4% for the standard cultivar and which guaranteed high quality of the beer obtained. The cultivar Caskadior 3 fulfills all the 15 criteria of European Convention for beer industry (Holland) and possess high quantity of the extract in the malt – over 81%. Caskadior 3 is the basic material for producing beer “Zagorka”.

Seed producing is high paying, because the cultivar is close pollinated and the foreign pollination is not possible.

Reference

Georgieva. V. 2002. My farma. N 568, p.5.

Maich, R. and Ordonez, A. 2003. Improved meiotic index in hexaploid triticale (x *Triticosecale Wittmack).*Cytologia, 68, 303 – 306.

Fig. 1. Plants bundle of cultivar Caskadior 3

Barley Genetics Newsletter 36:30-43 (2006)

New SSR markers for barley derived from the EST database

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Introduction

There are currently 1,196 microsatellite [or simple sequence repeat (SSR)] marker primer sets that have been developed for barley (from both genomic libraries and EST databases) of which 504 have been mapped (Saghai Maroof et al., 1994; Becker and Heun, 1995; Liu et al., 1996; Struss and Plieske, 1998; Ramsay et al., 2000; Pillen et al., 2000; Holton et al., 2002; Thiel et al., 2003; Li et al., 2003; Yu et al., 2005). Unfortunately, the available SSR markers provide uneven coverage of the barley genome and are concentrated near the centromeres. We have compared the available mapped SSRs and identified 62 BINs out of the total of 99 barley BINs (http://barleygenomics.wsu.edu/; http://rye.pw.usda.gov/cgi-bin/gbrowse/BarleyBinMaps) that have poor coverage. Although this represents, 63% of the barley genome, only 31% of the available SSRs map to these BINs. Additional SSR markers are needed to increase coverage in these BINs. Moreover, some of the SSR markers recently published are restricted from being used to develop new barley varieties, thus there is still a need for additional publicly available SSR markers that can be used without restrictions.

Materials and Methods

Barley ESTs used for primer development were selected by using either rice BAC or wheat EST sequences in a BLASTn search for publicly available barley ESTs with the low complexity filter turned off (BLASTn searches were completed between October, 2002 and October, 2004) (http://www.ncbi.nih.gov/BLAST/). Matches with e-values between 0 and 1e-2 were used. The resulting barley ESTs were processed through the Tandem repeats finder which measures the rate at which the actual EST sequence matches a perfect repeat sequence (Benson, 1999; http://tandem.bu.edu/trf/trf.submit.options.html). Primers were designed for SSRs with 85-100% matches to the perfect repeat sequence. Primer pairs were designed to flank SSR motifs using Primer3 software (Rozen and Skaletsky, 2000; http://frodo.wi.mit.edu/cgi-bin/primer3/primer3_www.cgi) and screened on a set of mapping parents to identify which population(s) were appropriate for mapping. Primer pairs were tested using two to four PCR protocols to identify a protocol that resulted in clear products (Tables 1 and 2). Products were separated on 6% polyacrylamide gels and visualized using silver staining.

Newly developed SSR markers were mapped on the appropriate mapping population(s) including: Steptoe x Morex (Kleinhofs et al., 1993), Chevron x M69 (Canci et al., 2003), Frederickson x Stander (Mesfin et al., 2003), or Atahualpa x M81 (unpublished). We used JoinMap 3.0 for map construction (Van Ooijen and Voorrips, 2001). Assignment to BINs was based on adjacent mapped markers that have been previously assigned to BINs (http://barleygenomics.wsu.edu/; http://rye.pw.usda.gov/cgi-bin/gbrowse/BarleyBinMaps). Wheat STS markers were mapped on the barley populations to increase the coverage of chromosome 3 (3H) (Liu and Anderson, 2003).

Results and Discussion

A total of 76 new markers were developed that produce between two to six alleles per locus among the twelve mapping parents: Atahualpa, M81, Chevron, M69, Frederickson, Stander, Harrington, OUH602, Hor211, Lacey, Steptoe and Morex (Table 3). Sixty of the markers were mapped using the Steptoe x Morex, Chevron x M69, Frederickson x Stander, and Atahualpa x M81populations (Figure 1 and Table 3). Of the 60 mapped markers, 41 (68%) have mapped to BIN positions that were previously identified as being poorly covered with the currently available SSR markers. These markers should provide additional tools for barley genetic mapping and marker-assisted selection.

Acknowledgements

The authors would like to thank Charles Gustus and Danielle Wojdyla for their assistance in STS and SSR marker data collection. This work was carried out in part using software provided by the University of Minnesota Supercomputing Center. This research was supported by U. S. Barley Genome Project.

References:

Becker, J., and M. Heun. 1995. Barley microsatellites: allele variation and mapping. Plant Mol. Biol. 27: 835-845.

Benson, G. 1999. Tandem repeats finder: a program to analyze DNA sequences. Nucleic Acids Research 27: 573-580.

Canci, P.C., L.M. Nduulu, R. Dill-Macky, G.J. Muehlbauer, D.C. Rasmusson, and K.P. Smith. 2003. Genetic relationship between kernel discoloration and grain protein concentration in barley. Crop Sci. 43: 1671-1679.

Holton, T.A., J.T. Christopher, L. McClure, N. Harker, and R.J. Henry. 2002. Identification and mapping of polymorphic SSr markers from expressed gene sequences of barley and wheat. Mol. Breeding 9: 63-71.

Kleinhofs, A., A. Kilian, M.A. Saghai Maroof, R.M. Biyashev, P. Hayes, F.Q. Chen, N. Lapitan, A. Fenwick, T.K. Blake, V. Kanazin, E. Ananiev, L. Dahleen, D. Kudrna, J. Bollinger, S.J. Knapp, B. Liu, M. Sorrells, M. Heun, J.D. Franckowiak, D. Hoffman, R. Skadsen, and B.J. Steffenson. 1993. A molecular, isozyme and morphological map of the barley (*Hordeum vulgare*) genome. Theor. Appl. Genet. 86: 705-712.

Li , J.Z., T.G. Sjakste, M.S. Röder and M.W. Ganal. 2003. Development and genetic mapping of 127 new microsatellite markers in barley. Theor. Appl. Genet. 107: 1021–1027.

Liu, S., and J.A. Anderson. 2003. Targeted molecular mapping of a major wheat QTL for Fusarium head blight resistance using wheat ESTs and synteny with rice. Genome 46: 817-823.

Liu, Z.-W., R.M. Biyashev and M.A. Saghai Maroof. 1996. Development of simple sequence repeat markers and their integration into a barley linkage map. Theor. Appl. Genet. 93: 869–876.

Mesfin, A., K.P. Smith, R. Dill-Macky, C.K. Evans, R Waugh, C.D. Gustus, and G.J. Muehlbauer. 2003. Quantitative trait loci for Fusarium head blight resistance in barley detected in a two-rowed by six-rowed population. Crop Sci. 43: 307-318.

Pillen, K., A. Binder, B. Kreuzkam, L. Ramsay, R. Waugh, J. Förster, J. Léon. 2000. Mapping new EMBL-derived barley microsatellites and their use in differentiating German barley cultivars. Theor. Appl. Genet. 101: 652-660.

Ramsay, L., M. Macaulay, S. degli Ivanissevich, K. MacLean, L. Cardle, J. Fuller, K. J. Edwards, S. Tuvesson, M. Morgante, A. Massari, E. Maestri, N. Marmiroli, T. Sjakste, M. Ganal, W. Powell, and R. Waugh. 2000. A Simple Sequence Repeat-Based Linkage Map of Barley. Genetics 156: 1997-2005.

Rozen, S., and H. Skaletsky. 2000. Primer3 on the WWW for general users and for biologist programmers. In: Bioinformatics Methods and Protocols: Methods in Molecular Biology. Eds Krawetz, S., and Misener, S. Humana. Totowa, NJ. pp 365-386.

Saghai Maroof, M.A., R.M. Biyashev, G.P. Yang, Q. Zhang and R.W. Allard. 1994. Extraordinarily polymorphic microsatellite DNA in barley: species diversity, chromosomal locations and population dynamics. Proc. Natl. Acad. Sci. USA 91: 5466–5470.

Struss, D., and J. Plieske. 1998. The use of microsatellite markers for detection of genetic diversity in barley populations. Theor. Appl. Genet. 97: 308-15.

Thiel, T, W. Michalek, R.K. Varshney, and A. Graner. 2003. Exploiting EST databases for the development and characterization of gene-derived SSR-markers in barley (*Hordeum vulgare* L.). Theor. Appl. Genet. 106: 411–422.

Van Ooijen, J.W., and R.E. Voorrips. 2001. JoinMap 3.0, Software for the calculation of genetic linkage maps. Plant Research International, Wageningen, the Netherlands.

Table 1. PCR recipes for UMB SSR markers

PCR recipe	A	B	C
dNTPs (1.25 mM each)	1.0	1.0	1.0
$MgCl_2^2$ (25 mM)	1.0	1.0	0.5
10 X buffer *	1.0	1.0	1.0
Forward Primer (5µM)	0.6	0.6	1.0
Reverse Primer (5µM)	0.6	0.6	1.0
Betaine (5 M)	0	2.0	0
DMSO (100%)	0	0.5	0
Taq polymerase*	0.075	0.075	0.075
H_2O	2.725	0.225	2.425
DNA (10 ng/µL)	3.0	3.0	3.0
Total	10.0	10.0	10.0

* 10 X buffer is provided with purchase of Taq polymerase

Table 2. PCR programs for UMB SSR markers

Stage	Temp. (°C)	Time (min.)	Cycles	Stage	Temp. (°C)	Time (min.)	Cycles
Program A*				Program F*			
Initial Hold	95	9:00	1	Initial Hold	-	-	-
Thermal Cycles-1	94 64 72	1:00 0:30 1:00	18	Pre-PCR	95 58 72	9:00 1:00 1:00	1
Thermal Cycles-2	94 55 72	1:00 1:00 1:00	30	Thermal Cycles	94 58 72	0:30 0:30 0:30	30
Ending Hold	72	5:00	1	Ending Hold	72	5:00	1
Final Hold	4	∞	1	Final Hold	4	∞	1
Program J*				Program L58			
Initial Hold	95	8:00	1	Initial Hold	95	9:00	1
Thermal Cycles	94 60 72	1:00 1:00 2:00	35	Thermal Cycles	94 58 72	0:10 0:20 0:45	30
Ending Hold	72	5:00	1	Ending Hold	72	2:30	1
Final Hold	4	∞	1	Final Hold	4	∞	1

* Programs A, F and J are as in Ramsay et al., 2000.

Table 3. PCR and mapping information for UMB markers.

Name	Chr.	BIN[1]	Population[2] Polymorphism	Population[2] Mapped	Quality Score[3]	Alleles[4]	PCR Program	PCR Recipe
UMB101	1 (7H)	11-12	A/M; F/S; H/O; H/L; S/M	A/M; F/S; S/M	2	3	L58	A
UMB102	1 (7H)	11-12	A/M; H/O; H/L; S/M	S/M	2.5	2	L58	A
UMB103	1 (7H)	13	A/M; F/S; H/O; H/L; S/M	A/M; F/S; S/M	2	4	L58	A
UMB104	1 (7H)	7-8	C/M; F/S; H/O; H/L; S/M	F/S; S/M	1.5	2	F	A
UMB105	1 (7H)	10-11	A/M; C/M; H/O	A/M; C/M	3	2	L58	A
UMB106	1 (7H)	**4-5**	C/M; H/L; S/M	C/M; S/M	2	2	L58	A
UMB107	1 (7H)	**4-5**	F/S; H/O	F/S	3	4	J	C
UMB108	1 (7H)	2	A/M; F/S; H/O; H/L; S/M	F/S; S/M	1	3	A	B
UMB201	2 (2H)	6-7	C/M; F/S; H/O; H/L; S/M	C/M; F/S; S/M	2.5	4	L58	A
UMB202	2 (2H)	6-7	A/M; C/M; F/S; H/O; S/M	A/M; C/M; F/S; S/M	2	2	F	A
UMB203	2 (2H)	4-5	C/M; H/L	C/M	2	2	J	C
UMB204	2 (2H)	7-8	F/S; H/O	F/S	2.5	2	L58	A
UMB205a	2 (2H)	4-5	C/M	C/M	1	2	J	C
UMB205b	2 (2H)	4-5	A/M; C/M; H/O	C/M	1	3	J	C
UMB206	2 (2H)	**2-3**	C/M; F/S; H/O; H/L; S/M	F/S; S/M	2	3	L58	B
UMB301	3 (3H)	**1-2**	C/M; F/S; H/L; S/M	C/M; F/S; S/M	3	3	F	A
UMB302	3 (3H)	**4-6**	A/M; C/M; F/S; H/O; S/M	A/M; C/M; F/S; S/M	1	4	L58	A
UMB303	3 (3H)	**4-6**	A/M; C/M; F/S; H/O; S/M	A/M; C/M; F/S; S/M	2	6	L58	A
UMB304	3 (3H)	**4-6**	A/M; C/M; F/S; H/L; S/M	A/M; C/M; S/M	1	4	J	C
UMB305	3 (3H)	**2-4**	A/M; H/O; H/L	A/M	1	2	F	A
UMB306	3 (3H)	**2-4**	A/M; H/O; H/L	A/M	1	3	F	A
UMB307	3 (3H)	**2-4**	A/M; H/O; H/L	A/M	1	3	L58	A
UMB308	3 (3H)	**1-2**	A/M; C/M; F/S; H/O; S/M	F/S; S/M	2.5	2	F	B
UMB309	3 (3H)	**4-6**	A/M; C/M; F/S; S/M	S/M	3	4	J	B
UMB310	3 (3H)	**4-6**	A/M; C/M; F/S; H/L; S/M	C/M; S/M	2	3	L58	B
UMB311	3 (3H)	**16**	CM; F/S; H/L	C/M	3	3	J	B
UMB401	4 (4H)	**4-5**	H/O; H/L; S/M	S/M	1	2	J	B
UMB402	4 (4H)	**2-3**	H/O; H/L; S/M	S/M	1	2	L58	A

Table 3 cont. PCR and mapping information for UMB markers.

Name	Chr.	BIN[1]	Population[2] Polymorphism	Mapped	Quality Score[3]	Alleles[4]	PCR Program	PCR Recipe
UMB403	4 (4H)	**4-5**	A/M; C/M; H/O; H/L; S/M	C/M; S/M	2	4	L58	B
UMB404	4 (4H)	5-6	C/M; H/O; S/M	C/M; S/M	1	2	J	B
UMB501a	5 (1H)	6	C/M; F/S	C/M; F/S	2	2	L58	B
UMB501b	5 (1H)	6	C/M; F/S	C/M; F/S	3	2	L58	A
UMB502	5 (1H)	**12-13**	A/M; F/S; H/O	A/M; F/S	1	3	L58	A
UMB503	5 (1H)	**2**	A/M; C/M; F/S; H/O; H/L; S/M	C/M; S/M	2	6	L58	A
UMB504	5 (1H)	**11-12**	A/M; F/S; H/O; H/L; S/M	F/S; S/M	2	4	L58	B
UMB505	5 (1H)	**2**	A/M; C/M; H/O; H/L; S/M	S/M	3	3	J	B
UMB506	5 (1H)	6	C/M; F/S; H/O; H/L	C/M; F/S	2	2	J	B
UMB507	5 (1H)	6	C/M; F/S; H/O; H/L	C/M; F/S	1	3	L58	B
UMB508	5 (1H)	**14**	A/M; C/M; F/S; H/O; H/L	C/M	2.5	3	L58	B
UMB601	6 (6H)	**13-14**	A/M; F/S; S/M	A/M; F/S; S/M	1	2	F	B
UMB602	6 (6H)	**11-13**	A/M; C/M; F/S; H/O; H/L	A/M; C/M; F/S	2	4	F	A
UMB603	6 (6H)	**13-14**	A/M; C/M; F/S; H/O; H/L; S/M	A/M; C/M; F/S; S/M	2	4	L58	A
UMB604	6 (6H)	7-9	A/M; C/M; F/S; H/O; H/L; S/M	C/M; F/S; S/M	1	5	L58	A
UMB605	6 (6H)	**14**	A/M; C/M; F/S; H/O; H/L	C/M; F/S	1	2	L58	B
UMB606	6(6H)	**1**	A/M; C/M; F/S; H/O; H/L; S/M	S/M	3	3	F	B
UMB701	7 (5H)	**9**	A/M; F/S; H/O	F/S	2	2	J	B
UMB702	7 (5H)	**10-11**	C/M; H/O; H/L	C/M	1	3	F	A
UMB703	7 (5H)	**7-8**	A/M; S/M	S/M	1.5	2	L58	A
UMB704	7 (5H)	2-4	A/M; F/S; H/O; H/L	A/M; F/S	1	4	L58	A
UMB705	7 (5H)	**5-6**	F/S; H/O	F/S	1.5	3	L58	A
UMB706	7 (5H)	**9**	A/M; C/M; H/O; H/L	A/M; C/M	1	2	L58	A
UMB707	7 (5H)	**6-7**	A/M; F/S; H/O; S/M	A/M; S/M	1	3	L58	A
UMB708	7 (5H)	**10-11**	A/M; H/O	A/M	1	2	F	A
UMB709	7 (5H)	**10-11**		A/M; H/O; S/M	S/M	2	2	F
UMB710	7 (5H)	**11-12**		S/M	S/M	2	2	L58
UMB711	7 (5H)	**11-12**		A/M; F/S; H/O;	F/S; S/M	2	3	L58

Table 3 cont. PCR and mapping information for UMB markers.

Name	Chr.	BIN[1]	Population[2] Polymorphism	Mapped	Quality Score[3]	Alleles[4]	PCR Program	PCR Recipe
UMB712	7 (5H)	**13-14**	A/M; F/S; H/O; H/L; S/M	F/S; S/M	2	3	F	B
UMB713	7 (5H)	**13-14**	A/M; F/S; H/O; S/M	S/M	2	4	L58	B
UMB714	7 (5H)	**13-14**	A/M; F/S; H/O; H/L; S/M	F/S	3	2	F	B
UMB715	7 (5H)	**13**	A/M; F/S; H/O	F/S	2	5	J	B
UMB001			H/L		2	3	L58	B
UMB002			H/O		3	2	F	A
UMB003			H/O		2	4	F	A
UMB004			H/O; H/L		3	2	J	C
UMB005			H/O		1.5	2	L58	A
UMB006			H/O		3	2	L58	A
UMB007			H/O; H/L		2	3	L58	A
UMB008			H/O; H/L		1	3	A	C
UMB009			H/O		2	3	L58	A
UMB010			A/M; H/O; H/L; S/M		2.5	3	F	A
UMB011			A/M; H/O		1	3	L58	B
UMB012			H/O		1	2	L58	B
UMB013			H/L		2	2	L58	B
UMB014			A/M; C/M; H/O; H/L		2	3	J	B
UMB015			C/M; H/O; H/L; S/M		3	5	F	B
UMB016			H/O		1	2	A	B

[1] Bold type indicates BINs with poor SSR marker coverage

[2] Mapping populations: A/M=Atahualpa x M81; C/M=Chevron x M69; F/S=Frederickson x Stander; H/O=Harrington x OUH602; H/L=Hor211 x Lacey; S/M=Steptoe x Morex

[3] Scale from 1-5 where 1=Very easy to score and 5=Very hard to score

[4] Number of Alleles based on the twelve mapping parents

Table 4. Design information for UMB primers.

Name	EST Accession[1]	Forward Primer	Reverse Primer
UMB101	BE421034	CGGGTTCCATTGAGAAGAAC	CACAAATACAGATGCCGCAC
UMB102	CB881209	TTGTGTTTGAGATATCCTGTACTTTTC	ACCTTTTGCCGGCTTTTATT
UMB103	BQ762328	TGCCCATGAAGCCTCTTTAC	GGAACGGAGGGAGTATTAAGC
UMB104	CB881555	GGAAAAATAAACTATTCAACATCCTG	CAGCGCATGTGTTCTCAGAT
UMB105	BJ485220	GCCCCTGGTAAGAACTCCAT	CTGGGAACCGTACAGTGTTG
UMB106	AL502019	AGCATAAAGCCGCAAAAGAA	GCGTCCTGATGAAGAGGTGT
UMB107	CB873957	ACGCACGGGCATTTGTACT	GCCTGCATCATTTGTTTGTG
UMB108	CV055381	TCAAGCTGCTGCATTGCT	AGCCCAAACCCTTTTGTTTT
UMB201	BJ480735	GCTCCTGAAAAGGACCTCAG	TCTCCGCCACCTACACATAG
UMB202	BG299528	GGTCGGCTCCCTCTTCTACT	CGAGCGACATGAGGAACAT
UMB203	CB873608	TTTCATTGCTGTGACGGATG	AGCCTCACCCGGACTACC
UMB204	BM371159	GAATCCTCGGCCTTCTCAAC	GCGGAGCTTGACCTCGAC
UMB205a	CB881957	CGGTCGTAGAACGGAATCAG	GCACTTCCACCACAAGAAGC
UMB205b	CB881957	CGGTCGTAGAACGGAATCAG	GCACTTCCACCACAAGAAGC
UMB206	CB863325	GCGCTAGCTATCCACACAAA	AACATTAAGGGCGACAAGGA
UMB301	AV944239	CTTCACATGTCTGGGAAAACA	GACATGTTGGAAGGTGGCTT
UMB302	CD663662	ACCACAGGTAACCTCGCAAC	AAAGTGCTGGGAGCTTGAAA
UMB303	BU979287	CACGAGGGATGCTCTTGAGT	TGTATATTTCAAGCTCCCAGCA
UMB304	BM443659	CTTCGCTTACCGCTTTCG	TTTCAAGCTCCCAGCACTTT
UMB305	BJ484842	CAGAGCGGGCTCAACGTA	ACTTGCTGTCATCCTTGCTG
UMB306	BJ467519	GCAGAGCTGGCTCAACGTA	TTCACTGATCGACCACTTGC
UMB307	BJ461914	CTGCAGAGCTGGCTAACGTA	TTCACTGATCGACCACTTGC
UMB308	CB859861	CCCCTCAGGTTGTTCATCAT	AGCAGCAGCAACAACAACAG
UMB309	BF265771	GCTCGACTTCGAGGACACC	ATTCTTGCGGAACGACCTC
UMB310	CB879994	CTCCCAGCACTTTCACCATC	CCGATGCTCTTGAGTCGTG
UMB311	CA592691	ATCCAGTTTCAGCCACCAAC	ACCGCAGTGATCAGTGACAA
UMB401	AL507067	CGTCTTCGTACTCGCCTCTC	ATCGAGATGCACTCCCTCAT
UMB402	BQ466542	TCGATCCATCCAAACATGAG	CGTGTCACGTGTGTGTGTGT

Table 4 cont. Ordering and design information for UMB primers.

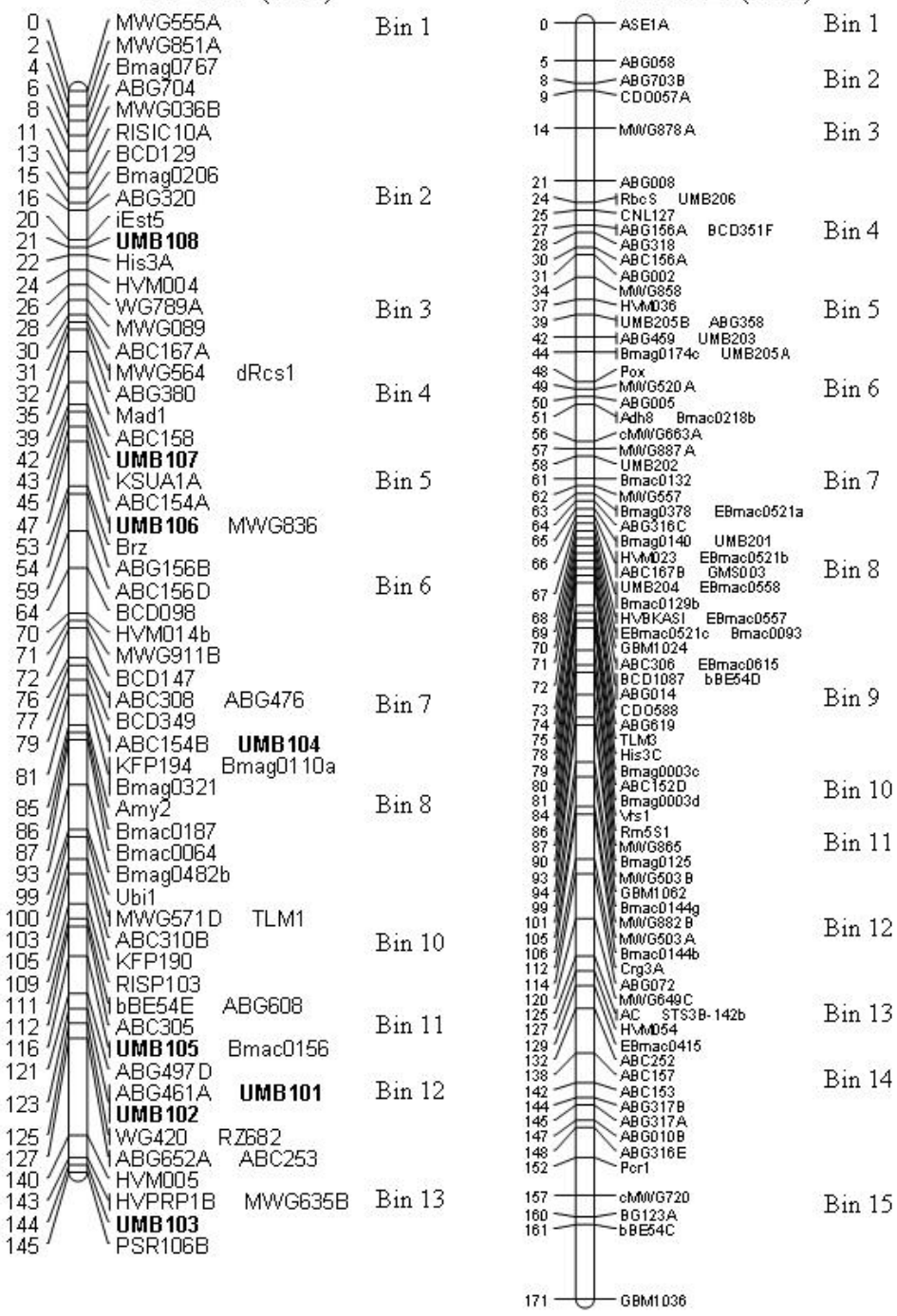
Name	EST Accession[1]	Forward Primer	Reverse Primer
UMB403	BM372825	TTCCGCAGATTCATTTCCAC	GCTGGACAGGCGTTAAAAAG
UMB404	AJ475924	GGAGGCAAGAACACTTGACAG	GCTCGATCTCCTCCTTGTTG
UMB501a	BE421033	CACACAGGCGACCATTTTC	CAGCTAGACGCTATGAGCCA
UMB501b	BE421033	CACACAGGCGACCATTTTC	CAGCTAGACGCTATGAGCCA
UMB502	BI953342	ATCCCATCTCCCTCCTCCTA	TGGAGTGCTCCTCCCAGTAG
UMB503	BQ766039	TCCCGGTGCCATATACAAAT	TTTGATGAAACGAAGGGAAA
UMB504	CV054443	CAAAGTGCGCGTGAGAAATA	AATCACCACCAGCTTCTTGG
UMB505	BE195848	ATGTTGCAGCAGAGCAGTTG	ATTGTTGGGGTTGTTCTTGC
UMB506	CD663377	CTCTTCCGTGAAACGAAACC	CGAGCAAGGACGTGGTAGAT
UMB507	CD663377	ATGTTTCAACAGGCCATTCC	CATGAAAACAGATGACGATGC
UMB508	BF621983	GATTAAGGCGTCCAATTCCA	TCGGGATGTGAAGAAGGAAC
UMB601	BJ486149	AAATACCGTATGGAGGGTGCT	CTACCCCTACGTCCGAGATG
UMB602	CB877685	AGGAGTGGGTCTCAGGTTTG	CAAGCAGATGCAACTACACCA
UMB603	CA006980	ATGAAACATCGCGAACTGTG	ACTGCAGTGAGGGAAGCTGT
UMB604	CA004840	GAGCAATCCCCTCATCCAAT	TCTTTGGTTTCCTCGTGTCC
UMB605	CD053629	GAGGCTTGTTCCTCAGACCA	ATGAGGAAGAGCGGGATCAG
UMB606	CV056304	CGAGCAGCAGCAGATCGT	CTCCTGCGCTTGGAGAAG
UMB701	BE421177	ACGTCGTGGATCAACGTGTA	TTACATTGCGCACAGCTAGG
UMB702	BF265777	CAGCATCCATCAGCAATGAA	CATGTTTGGCTTCTTCGTCC
UMB703	CA025623	GCCGCCTCTTACTCTTTGC	GGAGATGCCGAGGGACTT
UMB704	AV942720	ATCCTCCAACGAGGCACATA	GAGTCCATTTCACGGAGACC
UMB705	CB876579	TGCTGAGACACACACACACC	CGATGCACGAAAAGCTGTAG
UMB706	CB875298	TCAACAGATGACGTGCATGA	TCACACATTGAGGGAGGACA
UMB707	CB876579	TGCTGAGACACACACACACC	CGATGCACGAAAAGCTGTAG
UMB708	BE421505	CTCCTCAGCTCTGGAATGGA	GCGCATATACAAGCCAAACA
UMB709	CV063649	ACGACAAGACTATGGCAGCA	AAGGTTTCCTCAGCCTGTGA
UMB710	CV063745	ACAGTTCCCAACTTCCAAGC	CAGCACATCAGCCCGTACT
UMB711	BJ553047	GCAACGACACGTCTAAACCA	GTGGTGTGGTGTCCTTCCTC

Table 4 cont. Design information for UMB primers.

Name	EST Accession[1]	Forward Primer	Reverse Primer
UMB712	CB881537	CAAGAAGGAACGAAGCCAAG	TGCTAGTTTTTCCGGCTGAT
UMB713	CB881537	ATCAGCCGGAAAAACTAGCA	CAAGCGAAGAGGAAGAGGAA
UMB714	CB881537	GCAAGCAAATCACACTCTGG	GCGTCATCTACGGCTGATTC
UMB715	CB879524	CCGCGCCTAATTAACAAAAA	AGCTGACTGCTGACCAACCT
UMB001	BI949870	TTCTCCATGTTCGGCTTCTT	ATCAGCAATGAAGTTGTCGG
UMB002	BJ481660	GGCAAGTGAGCTCAACCTCT	GCCCTCACATGCAACATCTA
UMB003	CB881957	TCAGTGGTGACCGTGGTATC	GCACTTCCACCACAAGAAGC
UMB004	BG309433	ACCACGTCCACCACCATC	CTCTTTCTCCGCTCATCACC
UMB005	BQ763410	AAGAAAGCCCGGAAGAGAAG	ATGGTGCCATCCTGATTGAT
UMB006	BQ662872	CGGAGTCGCTTTCGAGATT	ACTGACAGCAACGGTGGTAG
UMB007	CB881851	CGCAATAAAATCGCAGGAAT	ACGGAAGACCGGGATAGTTT
UMB008	BU996005	CTGGAGCCTAGCTTGGAGTG	CTCCACCGTTCTTCACGTTT
UMB009	BI954579	CATCCCCATCCAGATCCA	GTCGAGAGGTGCGAGGATT
UMB010	BF621983	GCCACAGCCAAGAAAGCTAC	GATGGGATCTGCTTGGAGAG
UMB011	AV918630	TCTTCTCGCTAGCATCAGCA	CAAAGAAGGAGGTGGCTTCA
UMB012	BI960225	ATAGCGACGTGCTCCAGAGA	AGCAGCGACTTCCTTAGCAG
UMB013	CB873614	GAGCAAGCACGCACGTATTA	GGGACCTCGAGATGATCAAG
UMB014	CB882192	CAGGAGATCCGCGCTCTT	CGAGCAGTGAACGATGTACG
UMB015	CA030737	CGGACGAGGTTTACTCCAAA	AGCACAGGAGGATGAGGATG
UMB016	CA032410	CCATCACCCATTTCTTCCTC	GATGGATTTGTCCGTCCAAG

[1] EST accession from which the primers were designed (http://www.ncbi.nih.gov/)

Figure 1. Map locations of the UMB markers on consensus maps of the Steptoe x Morex (S/M), Frederickson x Stander (F/S) and Chevron x M69 (C/M) populations. Chromosome 7 (5H) is presented as a consensus map of the F/S and C/M populations with the S/M population separate. UMB508 (denoted with '✦') has had its location inferred from the C/M population (C/M is included in the consensus but the inferred marker did not map in the consensus). Markers denoted with '▶' have had their location inferred from the Atahualpa x M81 (A/M) population. Marker "ABG497D" [chr. 1 (7H), BIN 11], was named "ABG497B" in Canci et al. (2003), but the "ABG497B" locus should map to chr. 1 (7H), BIN 4, therefore we have designated a new locus name.


Chr. 1 (7H)
0 MWG555A
2 MWG851A
4 Bmag0767
6 ABG704
8 MWG036B
11 RISIC10A
13 BCD129
15 Bmag0206
16 ABG320
20 iEst5
21 UMB108
22 His3A
24 HVM004
26 WG789A
28 MWG089
30 ABC167A
31 MWG564 dRcs1
32 ABG380
35 Mad1
39 ABC158
42 UMB107
43 KSUA1A
45 ABC154A
47 UMB106 MWG836
53 Brz
54 ABG156B
59 ABC156D
64 BCD098
70 HVM014b
71 MWG911B
72 BCD147
76 ABC308 ABG476
77 BCD349
79 ABC154B UMB104
81 KFP194 Bmag0110a
Bmag0321
85 Amy2
86 Bmac0187
87 Bmac0064
93 Bmag0482b
99 Ubi1
100 MWG571D TLM1
103 ABC310B
105 KFP190
109 RISP103
111 bBE54E ABG608
112 ABC305
116 UMB105 Bmac0156
121 ABG497D
123 ABG461A UMB101
UMB102
125 WG420 RZ682
127 ABG652A ABC253
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Chr. 2 (2H)
0 ASE1A
5 ABG058
8 ABG703B
9 CDO057A
14 MWG878A
21 ABG008
24 RbcS UMB206
25 CNL127
27 ABG156A BCD351F
28 ABG318
30 ABC156A
31 ABG002
34 MWG858
37 HVM036
39 UMB205B ABG358
42 ABG459 UMB203
44 Bmag0174c UMB205A
48 Pox
49 MWG520A
50 ABG005
51 Adh8 Bmac0218b
56 cMWG663A
57 MWG887A
58 UMB202
61 Bmac0132
62 MWG557
63 Bmag0378 EBmac0521a
64 ABG316C
65 Bmag0140 UMB201
66 HVM023 EBmac0521b
ABC167B GMS003
67 UMB204 EBmac0558
Bmac0129b
68 HVBKASI EBmac0557
69 EBmac0521c Bmac0093
70 GBM1024
71 ABC306 EBmac0615
72 BCD1087 bBE54D
ABG014
73 CDO588
74 ABG619
75 TLM3
78 His3C
79 Bmag0003c
80 ABC152D
81 Bmag0003d
84 Vrs1
86 Rm5S1
87 MWG865
90 Bmag0125
93 MWG503B
94 GBM1062
99 Bmac0144g
101 MWG882B
105 MWG503A
106 Bmac0144b
112 Crg3A
114 ABG072
120 MWG649C
125 IAC STS3B-142b
127 HVM054
129 EBmac0415
132 ABC252
138 ABC157
142 ABC153
144 ABG317B
145 ABG317A
147 ABG010B
148 ABG316E
152 Pcr1
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160 BG123A
161 bBE54C
171 GBM1036
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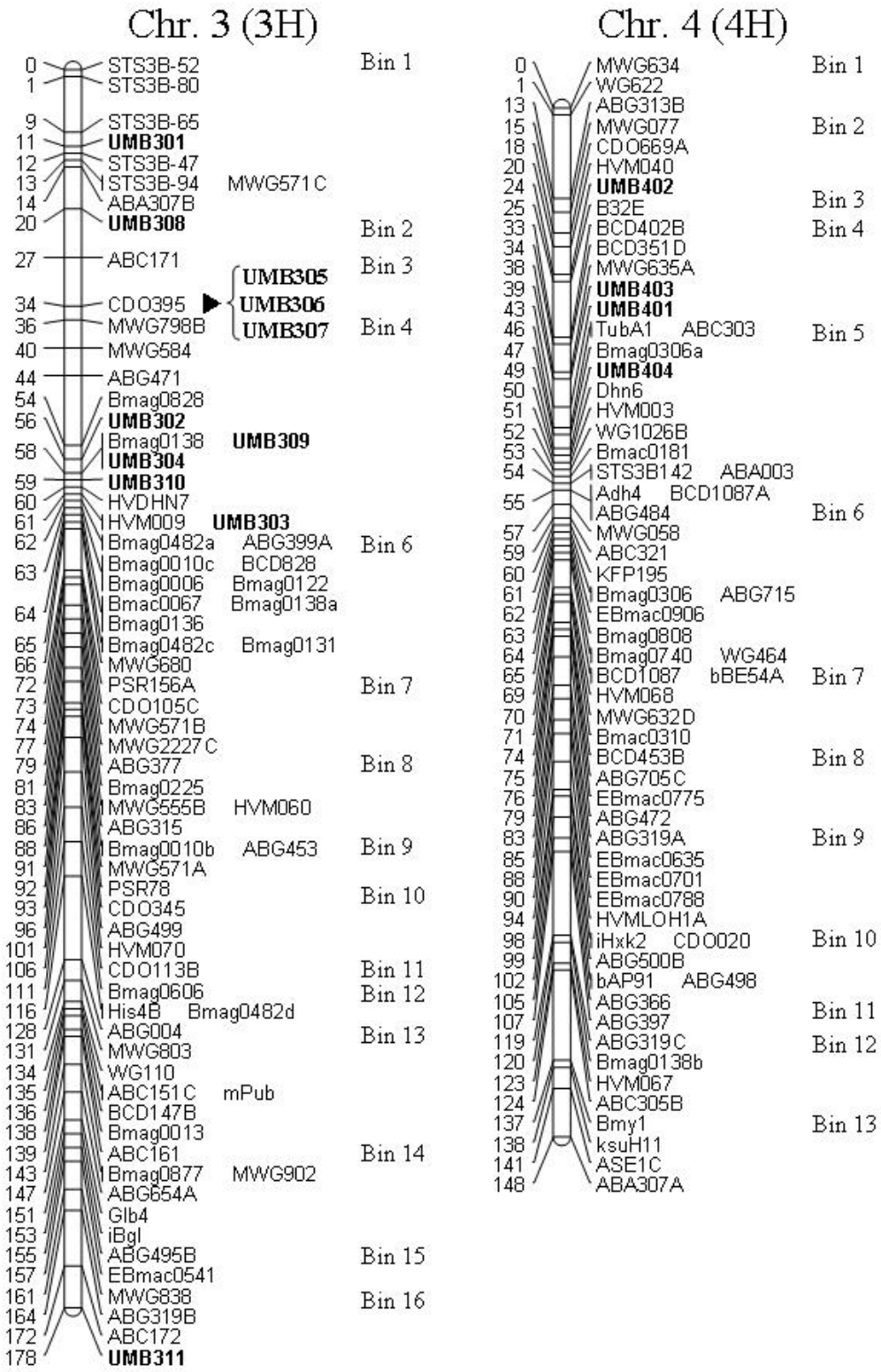
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9 STS3B-65
11 UMB301
12 STS3B-47
13 STS3B-94 MWG571C
14 ABA307B
20 UMB308
27 ABC171
34 CDO395
36 MWG798B
40 MWG584
44 ABG471
54 Bmag0828
56 UMB302
58 Bmag0138
UMB309
UMB304
59 UMB310
60 HVDHN7
61 HVM009 UMB303
62 Bmag0482a ABG399A
63 Bmag0010c BCD828
Bmag0006 Bmag0122
Bmac0067 Bmag0138a
64 Bmag0136
65 Bmag0482c Bmag0131
66 MWG680
72 PSR156A
73 CDO105C
74 MWG571B
77 MWG2227C
79 ABG377
81 Bmag0225
83 MWG555B HVM060
86 ABG315
88 Bmag0010b ABG453
91 MWG571A
92 PSR78
93 CDO345
96 ABG499
101 HVM070
106 CDO113B
111 Bmag0606
116 His4B Bmag0482d
128 ABG004
131 MWG803
134 WG110
135 ABC151C mPub
136 BCD147B
138 Bmag0013
139 ABC161
143 Bmag0877 MWG902
147 ABG654A
151 Glb4
153 iBgl
155 ABG495B
157 EBmac0541
161 MWG838
164 ABG319B
172 ABC172
178 UMB311
UMB305
UMB306
UMB307
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Chr. 4 (4H)
0 MWG634
1 WG622
13 ABG313B
15 MWG077
18 CDO669A
20 HVM040
24 UMB402
25 B32E
33 BCD402B
34 BCD351D
38 MWG635A
39 UMB403
43 UMB401
46 TubA1 ABC303
47 Bmag0306a
49 UMB404
50 Dhn6
51 HVM003
52 WG1026B
53 Bmac0181
54 STS3B142 ABA003
Adh4 BCD1087A
55 ABG484
57 MWG058
59 ABC321
60 KFP195
61 Bmag0306 ABG715
62 EBmac0906
63 Bmag0808
64 Bmag0740 WG464
65 BCD1087 bBE54A
69 HVM068
70 MWG632D
71 Bmac0310
74 BCD453B
75 ABG705C
76 EBmac0775
79 ABG472
83 ABG319A
85 EBmac0635
88 EBmac0701
90 EBmac0788
94 HVMLOH1A
98 iHxk2 CDO020
99 ABG500B
102 bAP91 ABG498
105 ABG366
107 ABG397
119 ABG319C
120 Bmag0138b
123 HVM067
124 ABC305B
137 Bmy1
138 ksuH11
141 ASE1C
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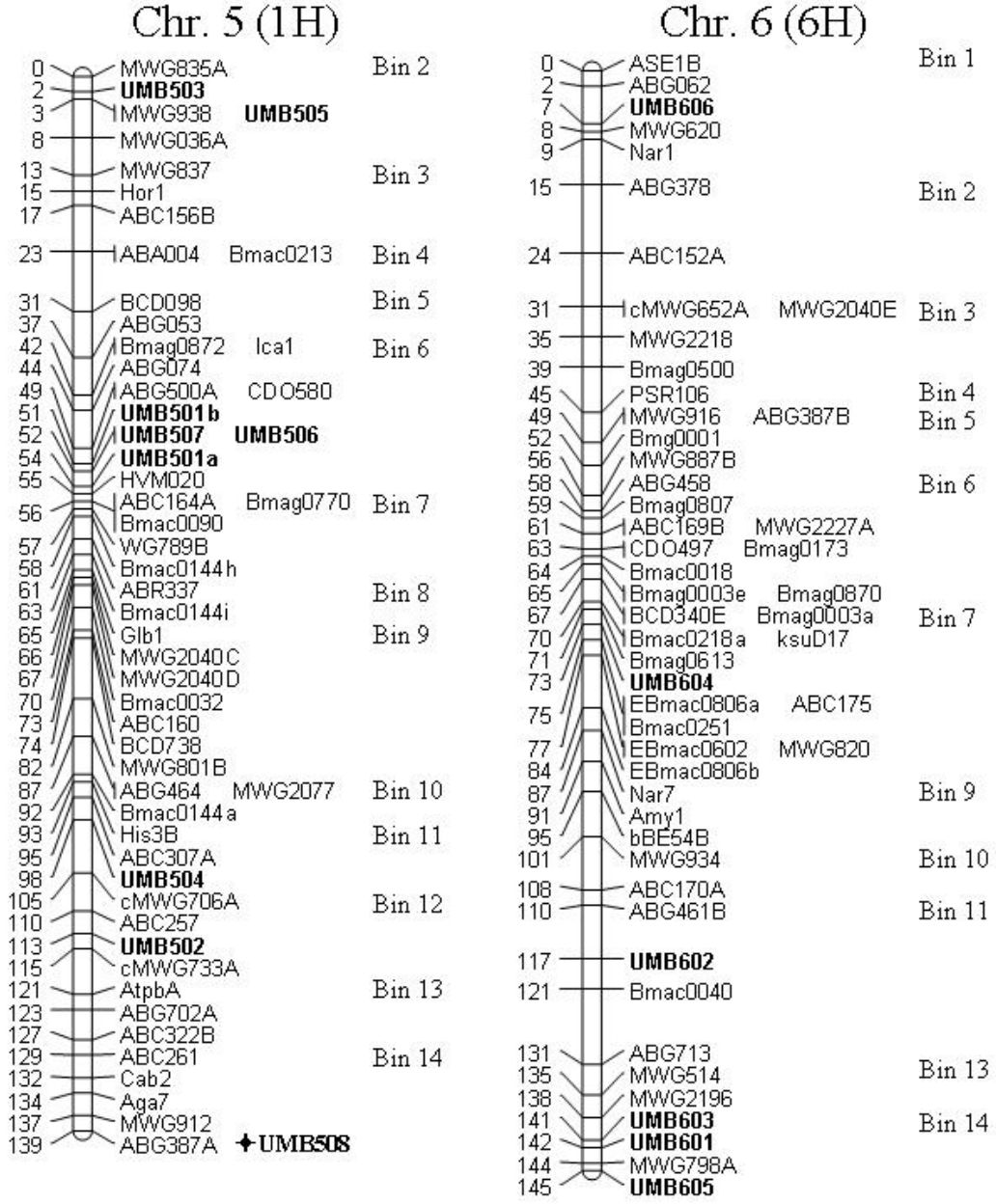
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23 ABA004 Bmac0213
31 BCD098
37 ABG053
42 Bmag0872 Ica1
44 ABG074
49 ABG500A CDO580
51 UMB501b
52 UMB507 UMB506
54 UMB501a
55 HVM020
56 ABC164A Bmag0770
Bmac0090
57 WG789B
58 Bmac0144h
61 ABR337
63 Bmac0144i
65 Glb1
66 MWG2040C
67 MWG2040D
70 Bmac0032
73 ABC160
74 BCD738
82 MWG801B
87 ABG464 MWG2077
92 Bmac0144a
93 His3B
95 ABC307A
98 UMB504
105 cMWG706A
110 ABC257
113 UMB502
115 cMWG733A
121 AtpbA
123 ABG702A
127 ABC322B
129 ABC261
132 Cab2
134 Aga7
137 MWG912
139 ABG387A ✦UMB508
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Chr. 6 (6H)
0 ASE1B
2 ABG062
7 UMB606
8 MWG620
9 Nar1
15 ABG378
24 ABC152A
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35 MWG2218
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87 Nar7
91 Amy1
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110 ABG461B
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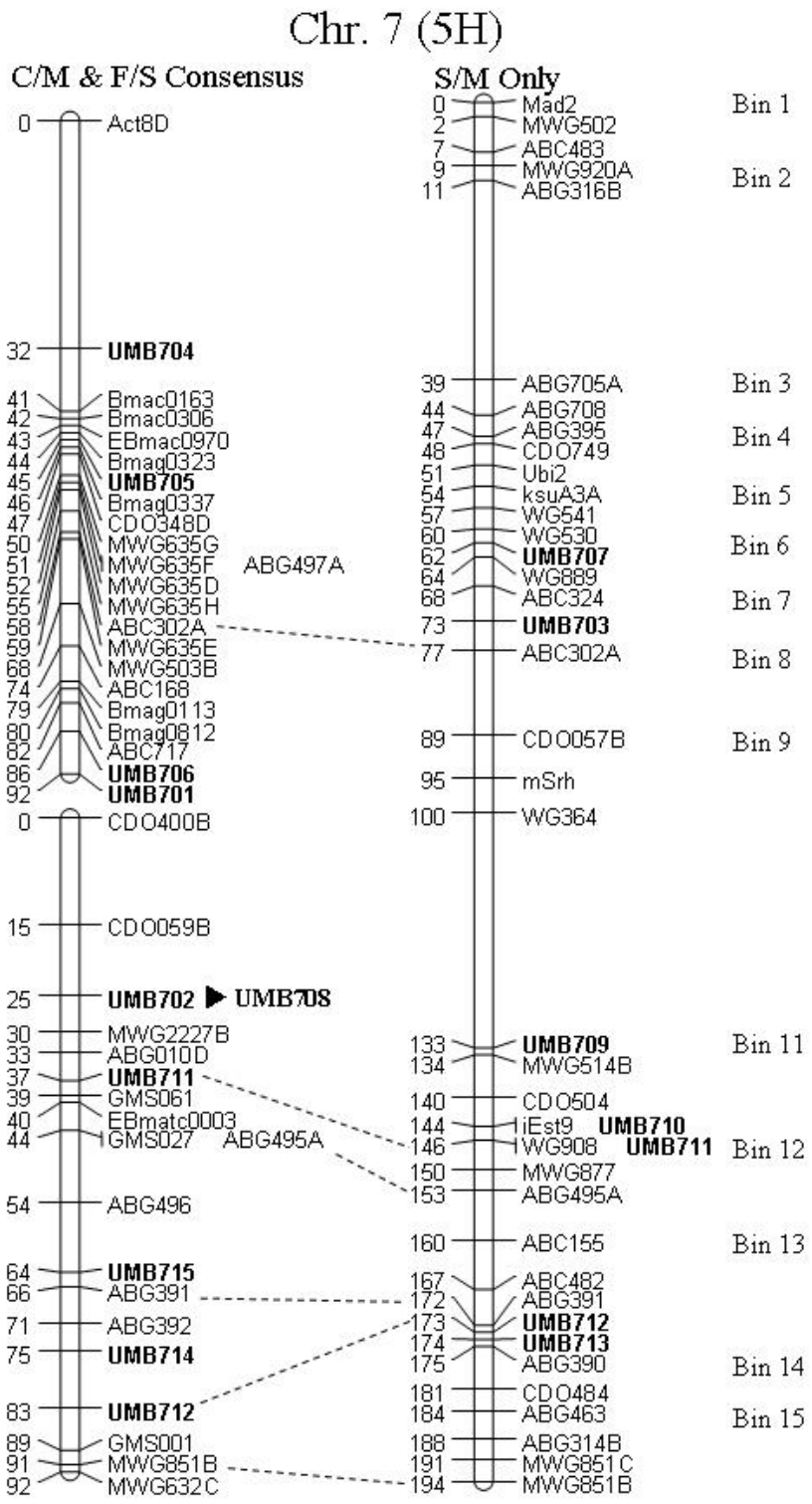
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44 Bmag0323
45 UMB705
46 Bmag0337
47 CDO348D
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51 MWG635F
52 MWG635D
55 MWG635H
58 ABC302A
59 MWG635E
68 MWG503B
74 ABC168
79 Bmag0113
80 Bmag0812
82 ABC717
86 UMB706
92 UMB701
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0 CDO400B
15 CDO059B
25 UMB702 ▶ UMB708
30 MWG2227B
33 ABG010D
37 UMB711
39 GMS061
40 EBmatc0003
44 GMS027
ABG495A
54 ABG496
64 UMB715
66 ABG391
71 ABG392
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51 Ubi2
54 ksuA3A
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77 ABC302A
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167 ABC482
172 ABG391
173 UMB712
174 UMB713
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Variability, correlation and Regression Analysis in Third Somaclonal Generation of Barley

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Abstract

Assessment of variability for qualitative and quantitative characters in twenty five SC_3 (somaclonal third generation) families of barley along with their mother cultivar Dissa was made. Two male sterile plants appearing like 'gigas' were obtained in one somaclonal family. Such plants had empty anther sac and spikelets remained open with wide angle for longer period. Segregation for two rowed and six rowed barley plants occurred in four somaclonal families where mother cultivar was six rowed. Considerable amount of variability existed among somaclonal families for plant height, tiller number, spike area, spikelet and grain number, 100 grain weight and grain yield per plant. Tiller number, grain number and 100 grain weight were the major characters for variation in grain yield in the population.

Key words*:* barley, SC_3 generation, qualitative and quantitative characters, somaclonal variation.

Introduction

Creation of genetic variability through tissue culture and its exploitation have become a major thrust for crop improvement programme. Somaclonal variation found among regenerated plants may not be stable because of physiological disturbances and epigenetic causes. In advance somaclonal generations, these nongenetic causes are eliminated and whatever variation is found is due to genetic changes. Somaclonal variation in advance generations are sufficiently stable and can be potentially used in crop improvement programme. Variation in morphological characters and *Helminthosporium* resistance in SC_2 generation and biochemical and cytological characters in SC_3 seeds of barley have been reported (Kole and Chawla, 1992; 1993). The present investigation was undertaken to study variability for qualitative and quantitative characters and to determine inter-relationships among yield and yield contributing characters in third somaclonal (SC_3) generation of barley.

Materials and Methods

The experimental materials comprised second generation seed progenies (SC_3 generation) obtained from 25 initial somaclones, regenerated from *in vitro* selected resistant calli against *Helminthosporium sativum*, derived from immature embryo explant of cultivar Dissa. The detailed procedures of regeneration are described earlier (Chawla and Wenzel, 1987). The 25 SC_3 families along with parent cv. Dissa were grown at Pant Nagar(29^oN, $79^o3'E$, 243.83msl), Uttaranchal, India during winter season in randomized block design with 3 replications. Each plot consisted of 5 rows of 2m length with intra- and inter-row spacings of 7.5 and 25cm. Plants

were carefully observed for changes in qualitative characters. Data were recorded on ten randomly selected plants from each plot in each replication for nine quantitative characters viz., plant height, tiller number per plant, days to heading, exertion of peduncle from flag leaf, spike area, spikelet number and grain number per spike, 100 grain weight and grain yield per plant. Estimates of phenotypic and genotypic coefficients of variation (Burton, 1952), heritability in broad sense and genetic advance (Johnson *et al*., 1955), correlation coefficient (Robinson *et al*., 1951) and regression analysis (Draper and Smith, 1981) were done following standard statistical methods.

Results and Discussion

Two plants in the progenies of one somaclonal family were quite distinct from early growth phase which appeared like *gigas*. Chromosome counts from root tip of SC_3 seeds of this family indicated these plants to be normal diploid. These plants were male sterile with empty anther sac and florets remained opened for longer time with wide angle. These male sterility is due to segregation of nuclear recessive gene(s), induced through the process of tissue culture.

Segregation for two rowed and six rowed barley plants occurred in the progenies of four somaclonal families where the parent variety Dissa was six rowed. Segregation of such somaclonal variants is due to genetic changes, induced through the process of tissue culture.

Among the quantitative characters studied mean squares due to somaclonal families were highly significant for all the characters studied, except days to heading and exertion of peduncle from flag leaf, indicating presence of somaclonal variation. Somaclonal variation for morphological traits has been reported by Dunwell *et al*. (1986) and Sozinov *et al*.(1988). The estimates of phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) were low for plant height and 100 grain weight; moderate for spike area and number of spikelets and grains per spike; and high for tiller number and grain yield per plant (Table 1). The estimates of heritability were low for plant height and spike area; moderate for tiller number and grains per spike and grain yield per plant; and high for spikelet number and 100 grain weight. Genetic advances were moderate for tiller number, spikelet number and grain yield per plant and low for rest of the characters. Moderate genetic advance coupled with moderately high to high heritability for spikelet number and grain yield indicates preponderance of additive gene action for these two characters. The results of heritability and genetic advance are in agreement Sinha and Saha (1999).

The results on genotypic and phenotypic correlations (Table 2) indicated that grain yield per plant had positive and significant correlations with tiller number, spikelet and grain number per spike and 100 grain weight at both genotypic and phenotypic levels. Similar results have been reported by Singh (1999). Among inter-character correlations, tiller number had positive and significant correlation with spikelet number, grain number and 100grain weight. Spikelet number also showed positive and significant correlation with 100 grain weight, although two rowed barley plants having less spikelet number had higher test weight. The overall results of correlation indicates the scope of selection of plants having higher tiller, spikelet and grain number with higher test weight which will be high yielding.

The stepwise multiple regression analysis following step up procedure indicated tiller number was the most important variable accounting 40.4% variability in grain yield. The other

two variables that were sequentially included in the regression equations were grain number per spike and 100 grain weight with the corresponding R^2 values of 0.52 and 0.60. This indicates that tiller number, grain number and grain weight jointly decided 60 % variation in grain yield. Therefore, selection of plants should be based on the above three characters for improvement in grain yield.

References

Burton, G.W. (1952): Quantitative inheritance in grasses. *Proc. 6th Int. Grassland Cong.* 11 : 277-283.

Chawla, H.S., Wenzel,G. (1987):*In vitro* selection of barley and wheat for resistance against *Helminthosporium sativum*. *Theor. Appl. Genet.* 74: 841-845.

Draper, N.R. , Smith, H. (1981): The step wise regression analysis procedure. In : *Applied Rgression Analysis (2nd ed.)* pp 307-313. John Wiley and Sons, New York.

Dunwell, J.M., Cornish,M., Powell,W., Bottino,E.M. (1986): An evaluation of field performance of the progeny of plants regenerated from embryos of Hordeum vulgare cv. Golden promise. *J. Agril. Sci.* 107: 561-564.

Johnson, H.W., Robinson, H.F., Comstock, R.E. (1955):. Estimates of genetic and environmental variability in soybean. *Agron. J.* 47: 477-483.

Kole, P.C., Chawla, H.S. (1992): Morphological and cytological variation in second somaclonal generation of barley. *Indian J. Genet.* 52: 114-117.

Kole, P.C., Chawla, H.S. (1993): Variation in *Helminthosporium* resistance and biochemical and cytological characteristics in somaclonal generations of barley. *Biol. Planta*, 35: 81-86.

Robinson, H.F., Comstock, R.E., Harvey, P.H. (1951): Genotypic and phenotypic correlations in corn and their implications in selection. *Agron. J.* 43 : 282-287.

Singh, B.P.(1999). Correlation study in barley (*Hordeum vulgare* L.):. *J. Appl. Biol.* 9:143-145.

Sinha,B.C. , Saha, B.C. (1999):. Genetic studies, heritability and genetic advance in barley (*Hordeum vulgare* L.). *J. Appl. Biol.* 9:108-116.

Sozinov, A.A. Iskakov, A.R., Gaponenko, A.K. (1988): Somatic cell culture – a new source of variation for barley breeding. *Dokl. Vsesoyuz. Akad. Sel'skokhoz. Nauk Im V.I. Lenina* 11: 3-5.

Table 1. Mean, range, phenotypic and genotypic coefficients of variation, heritability and genetic advance for seven quantitative Characters

Characters	Mean	Range		Coefficients of variation (%)		Heritability in broad sense (%)	Genetic advance as % of mean
		Min.	Max.	Phenotypic	Genotypic		
Plant height (cm)	81.57	75.03	98.00	9.27	5.58	36.3	6.93
Tiller number / plant	21.73	9.33	30.33	28.29	21.76	59.1	34.47
Spike area (cm2)	25.77	20.78	34.76	19.81	12.33	38.75	15.79
Spikelet No.	64.50	51.99	87.27	20.08	18.13	81.44	33.67
Grain No.	50.75	35.4	64.3	16.03	12.64	62.23	20.53
100 grain weight (g)	3.7	2.79	4.26	9.30	8.23	78.40	15.14
Grain yield per plant (g)	24.04	11.5	34.7	27.61	22.98	69.00	39.39

Table 2. Genotypic (G) and phenotypic (P) correlation coefficients among seven quantitative characters in SC_3 generation of barley

		Tiller No.	Spike area	Spikelet No.	Grain No.	100 grain weight	Grain yield per plant
Plant Height	G	0.527**	0.086	0.625**	0.021	0.111	0.194
	P	0.123	0.123	0.340**	0.038	0.028	0.034
Tiller No.	G		0.393**	0.268*	0.436**	0.247*	0.673**
	P		0.100	0.236*	0.342**	0.214	0.628**
Spike area	G			0.521**	0.891**	0.089	0.374**
	P			0.431**	0.495**	-0.009	0.179
Spikelet No.	G				0.525**	0.391**	0.365**
	P				0.452**	0.295**	0.308**
Grain No.	G					0.012	0.697**
	P					-0.104	0.532**
100 grain weight	G						0.458**
	P						0.396**

*, ** : Significant at P=0.05 and 0.01, respectively.

REPORTS OF THE COORDINATORS

Overall coordinator's report

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Since the latest overall coordinator's report in Barley Genetics Newsletter Volume 35, no changes of the coordinators have been reported. I do hope that most of you are willing to continue with this work and provide us with new important information and literature search in the future. Please observe some address changes have taken place since the last volume of BGN.

As it became decided at the 9th International Genetic Barley Symposium in Brno, 2004, the current system and trait coordination should continue but with a view towards whole genome coordination. Bill Thomas and Dave Marshall from the Scottish Crop Research Institute, Invergowrie, Dundee, UK, are investigating the potential of modernizing the overall system and integrating all types of current and historic data collections into a single, combined database. They are working on this subject.

In this connection I also want to call upon the barley community to pay attention on the AceDB database for 'Barley Genes and Barley Genetic Stocks'. It contains much information connected with images and is useful for barley research groups inducing barley mutants and looking for new characters. It gets updated continuously and some more images are added to the original version. Also the germplasm part is under revision. The searchable address is: www.untamo.net/bgs

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Coordinator's Report: Barley Chromosome 1H (5)

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American six-row malting barleys possess an effective and durable resistance against spot blotch. In the variety 'Morex' Steffenson *et al*. (Steffenson *et al*. 1996) had dissected his resistance into a seedlings resistant on chromosome 7H, one major QTL for adult plant resistance on chromosome 1H and one minor QTL for adult plant resistance on chromosome 7H. This was done in a doubled haploid population from the cross 'Steptoe' × 'Morex', and 'Morex' contributet all alleles for resistance. In order to confirm these resistance genes, 'Morex' resistance against spot blotch was investigated in the crosses 'Dicktoe' × 'Morex' and 'Harrington' × 'Morex' (Bilgic *et al*. 2005). Additionally, the experiment in the cross 'Steptoe' × 'Morex' was repeated. While the latter experiment confirmed the QTL found before, no QTL on chromosome 1H was detected in the other two crosses.

The localistion of QTLs for straw-quality characteristics of barley under drought stress was the aim of Grando *et al*. (2005). For this purpose 494 F_7 recombinant inbred lines were scored in two years and two locations for acid detergent fiber (ADF), neutral detergent fiber (NDF), voluntary intake (INT), lignin content (LIC), crude protein (CP) and digestible organic matter in dry matter (OMD). Additionally, in one environment, the percentages of blades, sheaths and stems, respectively (PCB, PCH, PCS) were measured. On chromosome 1H, eight QTLs were found: one for NDF, INT and CP, one for ADF and PCS, one for PCH, two for INT, one for LIC and NDF, one for CP and one for INT and ash content.

Peighambari *et al*. (2005) performed a QTL analysis in 72 doubled haploid lines from the cross Steptoe × Morex for several agronomical traits scored in two years. On chromosome 1H, four different QTLs were detected: one for number of seeds per spike, one for the date of spike inititiation, one for spikes per plant and thousand-seeds-weight and one for date of flowering and date of maturity.

In order to localize QTLs for different disease resistances, Yun *et al*. (2005) analysed 104 F_6-plants from a cross between the *spontaneum*-line OUH602 and the cultivar 'Harrington'. They phenotyped the lines for resistance against powdery mildew, leaf scald, Septoria speckled leaf blotch, net type net blotch and spot blotch. On the short arm of chromosome 1H, they detected one QTL for powdery mildew (at or nearby the position of the *Mla*-locus), one QTL for scald and one QTL for net type net blotch. While the allele conferring resistance for scald and powdery mildew originated from OUH602, 'Harrington' contributed the allele for resistance against net type net blotch.

In an advanced backcross population (BC_2DH) originating from a cross between the *spontaneum* line ISR42-8 and the variety 'Scarlett', von Korff *et al*. (2005) detected QTLs for different disease resistances. On chromosome 1H, they found a major QTL for resistance against powdery mildew, at or near by the *Mla*-locus. The alleles of the *spontaneum*-line reduced disease severity by 51.5%.

Hori *et al*. (2005) presented an alternative approach for advanced backcrosses. They produced both doubled haploid lines and BC_3F_2 lines from a same cross between the Japanese malting barley variety 'Haruna Nijo' and the *spontaneum*-line H605. The linkage map was calculated in the population of doubled haploids and subsequently a QTL analysis was done in both populations for agronomic and phenotypic traits. On the short arm of chromosome 1H, one QTL was found for kernel weight and the number of spikelets per ear in the BC_3F_2. On the long arm of the same chromosome, they detected a QTL for the number of spikelets per ear in the doubled haploids.

In an attempt to find QTL influencing 'none-parasitic leaf spots' (NPLS), Behn *et al*. (2005) analysed 536 DH lines from a cross between the NPLS tolerant barley line 'IPZ 24727' and the variety 'Krona' and compared them with results published before (Behn *et al*. 2004) from a cross with the same *ant* line and the variety 'Barke' (all spring barley varieties). On chromosome 1H, they found a minor QTL NPLS-tolerance in each of the crosses, but on different regions of the chromosome. Additionally, they detected three different QTLs for heading date and two QTLs for plant height on the same chromosome.

Yin *et al* (2005) looked for QTLs representing inputs for a ecophysiological phenology model predicting flowering time in the cross 'Apex' × 'Prisma': f_0 as the minimum number of days from sowing to flowering under optimal conditions,. θ_1 and θ_2 as the development stage for the start and the end of the photoperiod-sensitive phase, respectively, and δ as the parameter characterizing the photoperiod-sensitivity. On chromosome 1H, they found 3 different loci: one for the θ_1, one for f_0 and one for all four parameters.

By addition lines, Nasuda *et al*. (2005) localised totally 701 EST sequences to the 7 barley chromosomes. Seventy one were assigned to chromosome 1H.

Rostoks *et al*. (2005) presented an integrated map from three populations originating from the crosses 'Steptoe' × 'Morex', 'Lina' × HS92 and 'Oregon Wolfe Barley Dominant' × 'Oregon Wolfe Barley Recessive'. Beside 904 RFLP, SSR, and AFLP markers localized before, the map is enriched by 333 EST unigenes, localized by SNPs, InDels or SSRs within these genes. For many of these unigenes, up- or down-regulation under different stress conditions is presented as well as the localization of the respective homologues in rice. On chromosome 1H, 41 unigenes were localized.

References

Behn, A., L. Hartl, G. Schweizer, and G. Wenzel. 2004. QTL mapping for resistance against non-parasitic leaf spots in a spring barley doubled haploid population. Theor. Appl. Genet. 108(7): 1229-1235.

Behn, A., L. Hartl, G. Schweizer, and M. Baumer. 2005. Molecular mapping of QTLs for non-parasitic leaf spot resistance and comparison of half-sib DH populations in spring barley. Euphytica 141(3): 291-299.

Bilgic, H., B. J. Steffenson, and P. M. Hayes. 2005. Comprehensive genetic analyses reveal differential expression of spot blotch resistance in four populations of barley. Theor. Appl. Genet. 111(7): 1238-1250.

Grando, S., M. Baum, S. Ceccarelli, A. Goodchild, F. Jaby El-Haramein, A. Jahoor, and G. Backes. 2005. QTLS for straw quality characteristics identified in recombinant inbred lines of a *Hordeum vulgare* x *H. spontaneum* cross in a Mediterranean environment. Theor. Appl. Genet. 110(4): 688-695.

Hori, K., K. Sato, N. Nankaku, and K. Takeda. 2005. QTL analysis in recombinant chromosome substitution lines and doubled haploid lines derived from a cross between *Hordeum vulgare* ssp. *vulgare* and *Hordeum vulgare* ssp. *spontaneum*. Mol. Breeding 16(4): 295-311.

Korff, M. von, H. Wang, and J. Léon. 2005. AB-QTL analysis in spring barley. I. Detection of resistance genes against powdery mildew, leaf rust and scald introgressed from wild barley. Theor. Appl. Genet. 111(3): 583-590.

Nasuda, S., Y. Kikkawa, T. Ashida, K. Sato, A. K. M. R. Islam, K. Sato, and T. R. Endo. 2005. Chromosomal assignment and deletion mapping of barley EST markers. Genes & Genetic Systems 80(5): 357-366.

Peighambari, S. A., B. Y. Samadi, C. Nabipour, Gilles, and A. Sarrafi. 2005. QTL analysis for agronomic traits in a barley doubled haploids population grown in Iran. Plant Sci. 169(6): 1008-1013.

Rostoks, N., S. Mudie, L. Cardle, J. Russell, L. Ramsay, A. Booth, J. Svensson, S. Wanamaker, H. Walia, E. Rodriguez, P. Hedley, H. Liu, J. Morris, T. Close, D. Marshall, and R. Waugh. 2005. Genome-wide SNP discovery and linkage analysis in barley based on genes responsive to abiotic stress. Mol. Genet. Genom. 274(5): 527.

Steffenson, B. J., P. M. Hayes, and A. Kleinhofs. 1996. Genetics of seedling and adult plant resistance to net blotch (*Pyrenophora teres* f. *teres*) and spot blotch (*Cochliobolus sativus*) in barley. Theor. Appl. Genet. 92(5): 552-558.

Yin, X. Y., P. C. Struik, F. A. van Eeuwijk, P. Stam, and J. J. Tang. 2005. QTL analysis and QTL-based prediction of flowering phenology in recombinant inbred lines of barley. J. Exp. Bot. 56(413): 967-976.

Yun, S., L. Gyenis, P. Hayes, I. Matus, K. Smith, B. Steffenson, and G. Muehlbauer. 2005. Quantitative trait loci for multiple disease resistance in wild barley. Crop Sci. 45(6): 2563-2572.

Coordinator's report: Chromosome 2H (2)

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Gottwald *et al*. (2004) reported on an attempt to isolate the gene controlling a gibberellic-acid insensitive dwarf mutant in barley. The locus was named *sdw3* and is closely linked to RFLP marker MWG2287 on 2HS near the centromere. The gene symbols *gai* and *GA-ins* were used for the mutant in line Hv287 in earlier publications (Börner *et al*., 1999). This region of 2HS is orthologous with a highly conserved region on rice chromosome 7L. ESTs in this region were used to identify three putative GA-related ORFs in rice that might correspond to the *sdw3* locus (Gottwald *et al*., 2004).

Dahleen *et al*. (2005) studied 27 mutants from various sources that were placed in the brachytic (*brh*) group of semidwarf mutants. Based on allelism tests and molecular mapping studies using simple sequence repeat (SSR) markers, the mutants occurred at 18 different loci. Three of the brachytic mutants were located on chromosome 2H: *ert-t* (*brh3.y*), *brh4.j*, and *brh10.l*. Several mutants earlier identified as having a *brh3* phenotype were found to be allelic at the *ert-t* locus. Since the *ert-t* locus symbol was the symbol first published for this locus, it will be the recommended symbol. The *ert-t* locus was positioned near the tip of 2HS distal from SSR marker Bmac0134. The *brh4* locus was positioned near bin 9 of 2HL and *brh10* was position in bins 4 or 5 of 2HS (Dahleen *et al*., 2005).

Hori *et al*. (2005) mapped QTLs for resistance Fusarium head blight (FHB), incited primarily by *Fusarium graminearum*, using recombinant inbred lines (RILs) from a cross between a resistant two-rowed accession 'Russian 6' and a very susceptible six-rowed accession H.E.S. 4 from Afghanistan. Reactions to FHB were determined using a cut spike test where field grown spikes were harvested at anthesis and sprayed with a conidial suspension. The six-rowed spike 1 (*vrs1*) and closed flowering (*cly1*/*Cly2*) loci were mapped on 2HL. Two QTLs for FHB severity were detected on 2HL: one near the *vrs1* locus in bin 10 and one near the *cly1/Cly2* locus in bin 13. Rachis internode length was correlated with FHB severity. Other QTLs found on 2HL included early heading in bin 8, plant height and number fertile rachis nodes (spike length) in bin 10, and rachis internode length near bin 13.

Hori *et al*. (2006) used two-rowed barley accessions from China and Turkey to map QTLs for resistance to FHB. A set of recombinant inbred lines (RILs) was developed with 'Harbin' as the resistant parent and 'Turkey 6' as the susceptible parent. Using the cut spike to test FHB reactions, QTLs for FHB severity were not detected in the bin 7 to 10 region of 2HL. This result suggests that these two-rowed parents were homogeneous for QTLs controlling FHB severity in this region. A QTL for FHB severity was detected on 2HL and positioned near (5.8 cM) the closed flowering (*cly1*/*Cly2*) locus, probably in bin 13. Rachis internode length was correlated with FHB severity in this study.

Horsley *et al*. (2006) reported that chromosome 2HL contains a series of agronomically important traits and QTLs for resistance to FHB and for the accumulation of the toxin deoxynivalenol (DON). 'Foster', a Midwest six-rowed cultivar, was crossed to the resistant

two-rowed accession CIho 4196. RILs were evaluated in 10 field grown tests for FHB and in several tests for DON accumulation and for morphological traits. QTLs for various traits were found primarily on 2HL. QTLs for FHB severity and DON level were in bins 8 and 10 and were named *Qrgz-2H-8* and *Qrgz-2H-10*, respectively. These QTLs have been found in several other studies where FHB resistance was evaluated in crosses between two- and six-rowed cultivars. A QTL for DON was found in bin 2 of 4HS. A QTL for early heading was found in bin 8 of 2HL and is presumably the *Eam6* gene from the six-rowed parent. A QTL for low number of fertile rachis nodes was located in bin 10 near the six-rowed spike 1 (*vrs1*) locus. This QTL probably was identified earlier as the *lin1* locus. One or two QTLs for plant height were also found very close to the *vrs1* locus. Since the genes *Eam6*, *lin1*, and *vrs1* and the QTLs for susceptibility to FHB and shortness were all contributed by the six-rowed cultivar, breeding adapted lines with improved FHB resistance has been difficult in six-rowed barley. QTLs for spike angle and spike density or rachis internode length were located in bin 13 of 2HL. A number of these associations on 2HL were previous reported by Dahleen *et al*. (2003).

The transfer of favorable genes from wild barley to cultivated barley was evaluated in backcross two of a doubled-haploid population by von Korff *et al*. (2006). Early heading and short stature were associated with the early maturity 1 (*Eam1* or *Ppd-H1*) gene in the bin 3 region of 2HS. A second QTL for short stature was found in the bin 7 to 9 region of 2HL. A QTL for lodging resistance was found in bins 12 to 13 of 2HL.

Sameri and Komatsuda (2004) studied heading time in barley using RILs from a cross between a winter six-rowed accession and a spring two-rowed cultivar. Heading times for the RILs were estimated under long-day, short-day, and continuous light conditions. Two QTLs for early heading were detected on 2H under both spring and fall sown conditions, but not under continuous light. The QTL near the centromere from the winter parent, Azumamugi, probably corresponds to the *Eam6* or *eps2S* locus. The QTL on 2HL was also from the winter parent, but at a position not frequently associated with early maturity genes in barley.

Liu *et al*. (2005) identified in barley two full-length cDNA sequences homologous to caleosin, a seed-storage oil-body protein from sesame. The cDNAs, named *HvClo1* and *HvClo2*, are paralogs that cosegregate and were mapped on chromosome 2HL in bin 9 near marker CDO588.. *HvClo1* is expressed during late stages of embryogenesis and is seed specific. *HvClo2* is expressed in endosperm tissues during grain development.

Tondell *et al*. (2006) observed that four of twelve drought tolerance QTLs found on a barley consensus map were associated with regulatory candidate genes that mapped in similar genome positions. One of the four candidate genes is on chromosome 2HL in the bin 9 region.

Rostoks *et al*. (2005) used SNP discovery and linkage analysis to construct an integrated SNP map of more than 300 SNP loci. With the integration of RFLP, AFLP, and SSR markers, the map contained a total of 1,237 loci. Two regions of chromosome 2H were associated with QTLs for seedling tolerance to high salt concentrations.

References:

Börner, A., V. Korzun, S. Malyshev, and V. Ivandic. 1999. Molecular mapping of two dwarfing genes differing in their GA response on chromosome 2H of barley. Theor. Appl. Genet. 99:670-675.

Dahleen, L.S., H.A. Agrama, R.D. Horsley, B.J. Steffenson, P.B. Schwarz, A. Mesfin, and J.D. Franckowiak. 2003. Identification of QTLs associated with Fusarium head blight resistance in Zhedar 2. Theor. Appl. Genet. 108:95-104.

Dahleen, L.S., L.J. Vander Wal, and J.D. Franckowiak. 2005. Characterization and molecular mapping of genes determining semidwarfism in barley. J. Hered. 96:654-662.

Gottwald, S., N. Stein, A. Börner, T. Sasaki, and A. Graner. 2004. The gibberellic-acid insensitive dwarfing gene *sdw3* of barley is located on chromosome 2HS in a region that shows high colinearity with rice chromosome 7L. Mol. Gen. Genomics 271:426-436.

Hori, K., T. Kobayashi, K. Sato, and K. Takeda. 2005. QTL analysis of Fusarium head blight resistance using a high-density linkage map in barley. Theor. Appl. Genet. 111:1661-1672.

Hori, K., K. Sato, T. Kobayashi, and K. Takeda. 2006. QTL analysis of Fusarium head blight severity in recombinant inbred population derived from a cross between two-rowed barley varieties. Breed. Sci. 56:25-30.

Horsley, R.D., D. Schmierer, C. Maier, D. Kudrna, C.A. Urrea, B.J. Steffenson, P.B. Schwarz, J.D. Franckowiak, M.J. Green, B. Zhang, and A. Kleinhofs. 2006. Identification of QTLs associated with Fusarium head blight resistance in barley accession CIho 4196. Crop Sci. 46:145-156.

Korff, M. von, H. Wang, J. Léon, and K. Pilen. 2006. AB-QTL analysis in spring barley: II. Detection of favourable exotic alleles for agronomic traits introgressed from wild barley (*Hordeum vulgare* ssp. *spontaneum*). Thoer. Appl. Genet. 112:1221-1231.

Lui H., P. Hedley, L. Cardle, K.M. Wright, I. Hein, D. Marshall, and R. Waugh. 2005. Characterization and functional analysis of two barley caleosins expressed during barley caryopsis development. Planta 221:513-532.

Rostoks, N., S. Mudie, L. Cardle, J. Russell, L. Ramsay, A. Booth, J. T. Svensson, S.I. Wanamaker, H. Walia, E.M. Rodriguez, P.E. Hedley, H. Liu, J. Morris, T,J. Close, D.F. Marshall, and R. Waugh. 2005. Genome-wide SNP discovery and linkage analysis in barley based on genes responsive to abiotic stress. Mol. Gen. Genomics 274:515-527.

Sameri, M. and T. Komatsuda. 2004. Identification of quantitative trait loci (QTLs) controlling heading time in the population generated from a cross between Oriental and Occidental barley cultivars (*Hordeum vulgare* L.). Breed. Sci. 54:327-332.

Tondell, A. E. Francia, D. Barabschi, A. Aprile, J.S. Skinner, E.J. Stockinger, A.M. Stanca, and N. Pecchioni. 2006. Mapping regulatory genes as candidates for cold and drought stress tolerance in barley. Theor. Appl. Genet. 112:445-454.

Coordinator's Report: Barley Chromosome 3H

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Over the last year there have been a number of publications reporting the mapping of genes and QTL on barley chromosome 3H. The largest number of genes assigned to 3H was the 271 mapped by Cho *et al.* (2006) using transcriptome analysis on the wheat-barley disomic chromosome addition lines. Rostoks *et al*. (2005) mapped 51 genes to 3H as part of a genome–wide SNP discovery programme in which over 300 genes responsive to abiotic stress were mapped, mostly as SNPs. These publications confirmed the close syntenic relationship between barley 3H and rice chromosome 1. This synteny was used by Mammadov *et al*. (2005) to direct the development of 9 EST-derived STS markers that were mapped onto a high resolution map of the leaf rust resistance gene *Rph5* region on 3HS including five that co-segregated with the resistance gene. Hori *et al*. (2005) also published mapping data based on 60 EST derived markers, 7 of which mapped to 3H. These represent a small subset of 163 mapped to 3H by Sato *et al*. (2004), however primer information on the seven is given in Hori *et al*. (2005) allowing the association of EST sequences to the loci.

The mapping of individual genes has also reported in the last year with the barley homologue of GIGANTEA, *HvGI*, mapping to a syntenic position on 3HS (Bin 5-6) (Dunford *et al*., 2005). This gene is the homologue of an *Arabidopsis* flowering time regulator, however its map position does not correspond to the map position of any known flowering time QTL in barley. Skinner *et al*. (2006) reported the mapping of *HvICE2* a homologue of an Arabidopsis low temperature regulatory gene to 3HL (Bin 13-14). However, again, the map position of this candidate gene did not correspond to the position of a known low-temperature tolerance QTL.

The barley homologue of *acsF,* an enzyme involved in chlorophyll biosynthesis, was mapped to the short arm of chromosome 3H through the use of the wheat-barley disomic chromosome addition lines and was shown to be the known mutant *Xantha-l* (Rzeznicka *et al*.,. 2005).

Although much mapping work utilised the growing genomic resources in barley there were reports that used more generic approaches. Thus Mammadov *et al*. (2006) utilised degenerate primers designed to conserved motifs of the NBS region in known resistance genes to isolate 190 resistance gene analogues (RGA) clones from barley genomic DNA and mapped two of them to 3H (Bin 4 and Bin 14) using the Steptoe x Morex DH mapping population. AFLP have been used for detailed mapping of the *btr1/btr2* locus on 3HS (Senthil and Komatsuda, 2005) and some of these have been converted to STS markers (Azhacuvel *et al*., 2006).

Again this year a considerable number of QTL were reported in the literature some of which mapped to 3H. These included an increasing number of reports using recombinant chromosome substation lines to delineate association of quantitative traits with genomic regions (Hori *et al*., 2005, von Korff *et al*., 2005, 2006, Yun *et al*., 2006). Thus von Korff *et al*. (2005) report QTL for powdery mildew resistance on 3HS (Bin 5-6) and on 3HL (Bin 13-15) with the latter interval also housing QTL for resistance to leaf rust and scald. The same population, derived from *H. vulgare spontaneum* introgressions into the spring barley cultivar

Scarlett, has also been assessed for agronomic traits (von Korff *et al*., 2006). Several traits are reported to be associated with regions on chromosome 3H including brittleness of the rachis with a region on 3HS (Bin 3-6) and a large number of traits including height and harvest index with a region on 3HL (Bin 10-16). The authors postulate that these associations could be explained by the segregation of *btr1* and *sdw-1* (denso) respectively in this population. Other QTLs on 3H found in this study do not have obvious candidate genes but are consistent with other studies. Thus a QTL for thousand grain weight found on the distal end of 3HL (Bins 14-15) appears to relate to a similar QTL found by Hori *et al*. (1995) in a doubled haploid population derived from a cross between the cultivar Haruna Nijo and a *Hordeum sponteneum* accession. Other QTL found on populations derived from the same cross include ear length, number of spikelets and culm length (Hori *et al*., 1995).

Other studies that report QTL on 3H include those for agronomic characters discovered using the Steptoe x Morex mapping population reported by Peighambari *et al*. (2005). The QTL found on 3H include those for date of flowering, date of maturity, plant height and spike length (Peighambari *et al*., 2005). In an extensive study on straw quality characteristics reported by Grando *et al*. (2005) the QTL reported on 3H include those for acid detergent fibre, lignin content, voluntary intake and digestible organic matter (Grando *et al*., 2005).

In addition to the work reported in von Korff *et al*. (2005) other disease resistance QTL have been reported on 3H in the last year. Bilgic *et al*. (2005) found a total of four QTL for seedling (Bins 4-6 and 11-12) and adult resistance (Bins 2-4 and 9-11) to spot blotch in a study comparing resistance expression in four populations. The authors postulate that the seedling and adult resistances could relate to the same underlying QTL and note that the resistance mapped to 3HS does not correspond to anything reported previously (Bilgic *et al*., 2005). Yun *et al*. (2005) report a net blotch QTL on 3H (Bin 6) shown in a RIL population derived from a cross between H. *vulgare spontaneum* (OUH602) and the cultivar Harrington. This QTL was confirmed in a RCSL population derived from the same cross (Yun *et al*., 2006).

References:

Azhacuvel P., D. Vidya-Saraswathi, and T. Komatsuda. 2006. High-resolution linkage mapping for the non-brittle rachis locus *btr1* in cultivated x wild barley (*Hordeum vulgare*) Plant Sci 170: 1087-1094.

Bilgic H, B. Steffenson, and P. Hayes. 2005. Comprehensive genetic analyses reveal differential expression of spot blotch resistance in four populations of barley. Theor Appl Genet 111: 1238-1250.

Cho, S.H., D.F. Garvin, and G.J. Muehlbauer. 2006. Transcriptome analysis and physical mapping of barley genes in wheat-barley chromosome addition lines. Genetics 172: 1277-1285.

Dunford, RP., S. Griffiths, V. Christodoulou, and D.A. Laurie. 2005. Characterisation of a barley (*Hordeum vulgare* L.) homologue of the *Arabidopsis* flowering time regulator *GIGANTEA*. Theor Appl Genet 110: 925-931.

Grando S, M. Baum, S. Ceccarelli, A. Goodchild, F.J. El-Haramein, A. Jahoor, and G. Backes. 2005. QTLs for straw quality characteristics identified in recombinant inbred lines of a *Hordeum vulga*re x *H spontaneum* cross in a Mediterranean environment Theor Appl Genet 110 : 688-695

Hori, K., K. Sato, N. Nankaku, and K. Takeda. 2005. QTL analysis in recombinant chromosome substitution lines and doubled haploid lines derived from a cross between *Hordeum vulgare* ssp *vulgare* and *Hordeum vulgare* ssp *spontaneum*. Mol Breeding 16:295-311.

Korff M. von, H. Wang, J. Léon, and K. Pillen K.. 2005. AB-QTL analysis in spring barley. I. Detection of resistance genes against powdery mildew, leaf rust and scald introgressed from wild barley. Theor Appl Genet 111: 583-590.

Korff M. von, H. Wang, J. Léon, and K. Pillen. 2006. AB-QTL analysis in spring barley. II. Detection of favourable exotic alleles for agronomic traits introgressed from wild barley. Theor Appl Genet 112: 1221-1231.

Mammadov, J.A., Z. Liu, R.M. Biyashev, G.J. Muehlbauer, and M.A.S. Maroof.. 2006. Cloning, genetic and physical mapping of resistance gene analogs in barley (*Hordeum vulgare* L.). Plant Breeding 125: 32-42.

Mammadov, J.A., B.J. Steffenson, and M.A.S. Maroof. 2005. High-resolution mapping of the barley leaf rust resistance gene *Rph5* using barley expressed sequence tags (ESTs) and synteny with rice. Theor Appl Genet 111: 1651-1660.

Peighambari, S.A., B.Y. Samadi, A. Nabipour, G. Charmet and A. Sarrafi. 2005. QTL analysis for agronomic traits in a barley doubled haploids population grown in Iran. Plant Sci 169: 1008-1013.

Rostoks, N., S. Mudie, L. Cardle, J. Russell, L. Ramsay, A. Booth, J.T. Svensson, S.I. Wanamaker, H. Walia, E.M. Rodriguez, P.E. Hedley, H. Liu, J. Morris, T.J. Close, D.F. Marshall, and R. Waugh. 2005. Genome-wide SNP discovery and linkage analysis in barley based on genes responsive to abiotic stress. Mol Gen Genom 274: 515-527.

Rzeznicka, K., C.J. Walker, T. Westergren, C.G. Kannangara, D. von Wettstein, S. Merchant, S.P. Gough and M.Hansson. 2005. *Xantha-l* encodes a membrane subunit of the aerobic Mg-protoporphyrin IX monomethyl ester cyclase involved in chlorophyll biosynthesis. Proc Nat Acad U.S.A. 102: 5886-5891.

Sato, K., N. Nankaku, Y. Motoi, and K. Takeda. 2004. A large scale mapping of ESTs on barley genome. *In*: J. Spunar and J, Janikova (eds.), pp. 79-85. Barley Genetics IX. Proc.Ninth Int. Barley Genet. Symp., Brno, Czech Republic, June 20-26 2004.

Senthil, N. and T. Komatsuda. 2005. Inter-subspecific maps of non-brittle rachis genes *btr1/btr2* using occidental, oriental and wild barley lines. Euphytica 145: 215-220.

Skinner J., P. Szűcs, J. von Zitzewitz, L. Marquez-Cedillo, T. Filichkin, E.J, Stockinger, M.F. Thomashow, T.H.H. Chen, and P.M. Hayes. 2006. Mapping of barley homologs to genes that regulate low temperature tolerance in *Arabidopsis*. Theor Appl Genet 112: 832-842.

Yun S.J, L. Gyenis, P.M. Hayes, I. Matus, K.P. Smith, B.J. Steffenson, and G.J. Muehlbauer. 2005. Quantitative trait loci for multiple disease resistance in wild barley. Crop Sci 45: 2563-2572.

Yun S.J, L. Gyenis, E. Bossolini, P.M. Hayes, I. Matus, K.P. Smith, B.J. Steffenson, R. Tuberosa, and G.J. Muehlbauer 2006. Validation of quantitative trait loci for multiple disease resistance in barley using advanced backcross lines developed with a wild barley. Crop Sci 46: 1179-1186.

Coordinators Report: Chromosome 5H(7)

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Winterhardiness in winter barley is controlled by regulatory elements of photoperiod sensitivity and vernalization response combined with the physical trait of low temperature tolerance. Of the six photoreceptors mapped on two mapping populations, only one, *HvPhyC*, coincided with a photoperiod response QLT on chromosome 5HL (Szucs *et al*., 2006). The vernalization locus *VRN-H1* (*HvBM5A*) whose expression is regulated by photoperiod has been mapped on chromosome 5HL and is closely linked to *HvPhyC*.

Reproductive frost tolerance is the ability of reproductive organs to tolerate low temperatures. A QTL on chromosome 5H for tolerance to frost-induced floret sterility and frost-induced grain damage was identified in three mapping populations (Reinheimer *et al.*, 2004). This locus is located close to the vrn-H1 locus on chromosome 5H and has been associated with the locus giving a response at both vegetative and reproductive developmental stages.

Seed dormancy is an important trait that can prevent preharvest sprouting and regulate germination during the malting process. A major seed dormancy QLT was detected on chromosome 5H plus two others on chromosome 1H in a mapping population derived from crossing the Japanese malting cultivar Haruna Nijo x H602 (*H. spontaneum – dormant*) (Sato *et al*., 2006). Seven EST markers were localized in the vicinity of the QLT on chromosome 5H.

Identification of QTL resistance to Fusarium Head Blight (FHB) continues to be a challenging exercise. Chromosome 5H appears to be a lesser contributor of FHB resistance QTL. For example, in recombinant inbred populations derived from two-rowed crosses of Harbin (R) x Turkey 6 (HR), resistance QTL were located on all chromosomes except 5H (Hori *et al*., 2006). However, in an RI population derived from Russia 6 (HR) x HES4 (HS), which was mapped with 1,255 markers, two pulative resistance loci were located on chromosome 2H and one on 5H (Takeda, 2004).

Of more general interest to barley geneticists are the assembly of a high density microsatellite consensus map and the sequencing of the barley chloroplast genome. The consensus microsatellite or SSR map was assembled by combining the information from six independent mapping populations. It consists of 784 unique microsatellite loci from 696 primers spanning 1,137.6 cM with an average density of one SSR marker every 1.45 cM (Varshney *et al*., 2006).

The chloroplast genome of barley consists of 136,462 bp, including a large single copy region of 80,600 bp, a small single copy region of 12,704 bp, plus a pair of inverted repeats of 21,597 bp. The genome consists of 104 genes, including 70 peptide-encoding genes, plus 30 tRNA and 4 rRNA genes that are duplicated in the inverted repeat (Saski et al., 2006). This genome is practically identical to other cereal chloroplast genomes, indicating that such genomes are highly conserved.

References:

Hori, K., K. Sato, and K. Takeda. 2006. Comparison of Fusarium heat blight resistance loci in barley RI populations. Poster 327. Plant and Animal Genome XIV Conference. (http://www.intl-pag.org/14/abstracts/PAG14_P327.htm).

Reinheimer, J.L., A.R. Bar, and J.K. Eglinton. 2004. QTL mapping of chromosomal regions conferring reproductive frost tolerance in barley (*Hordeum vulgareL.*). Theor. Appl. Genet. 109:1267-1274.

Saski, C.A., S.B. Lee, H. Daniell, and J.P. Tomkins. 2006. The complete DNA sequence of the sorghum and barley chloroplast genomes. Poster 235. Plant and Animal Genome XIV Conference. (http://www.intl-pag.org/14/abstracts/PAG14_P327.htm).

Sato, K., K. Hori, and K. Takeda. 2006. QTL for seed dormancy from wild barley *Hordeum Vulgare ssp. Spontaneum*. Poster 309. Plant and Animal Genomes XIV Conference. (http://www.intl-pag.org/14/abstracts/PAG14_P327.htm).

Szucs, P., I. Karsai, J. von Zitzewitz, L.D.D. Cooper, T.H.H. Chen, P.M. Hayes, and J.S. Skinner. 2006. Positional relationship between photoperiod response QTL and photoreceptor and vernalization genes in barley. Poster 351. Plant and Animal Genomes XIV Conference. (http://www.intl-pag.org/14/abstracts/PAG14_P327.htm).

Takeda, K. 2004. Inheritance of the Fusarium Head Blight resistance in barley. Czech J. Genet. Plant Breed. 40:143.

Varshney, R.K., T.C. Marcell, L. Ramsey, J. Russell, M.S. Roeder, N. Stein, P. Langridge, R. Waugh, R. Niks, and A. Graner. 2006. A high density microsatellite consensus map of barley. Workshop 304. Plant and Animal Genome XIV Conference. (http://www.intl-pag.org/14/abstracts/PAG14_P327.htm).

Coordinator's Report: Chromosome 7H

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2005 brought many reports of various QTLs detected in populations derived from wild x cultivated barley crosses, with the goal of transferring desirable genes into elite breeding lines. Hori *et al.* (2005b) located QTLs for glume length, rachis-internode length, dormancy after five and ten weeks, ear length and kernel weight on chromosome 7H in a population derived from a cross with *H. vulgare* ssp. *spontaneum* (accession H602). An examination of straw quality QTLs (Grando *et al*. 2005) located several loci on chromosome 7H, for traits acid detergent fiber, lignin content, voluntary intake, and percentage of sheaths by weight of the air-dried straw sample. They used a RIL population derived from *H. vulgare* ssp. *spontaneum* accession 41-1. Li *et al*. (2005) determined QTLs for yield, yield components and malting quality in an advanced backcross population with *H. vulgare* ssp. *spontaneum* accession HS213. Five QTLs were located on chromosome 7H, for heading date, ear length, spikelet number per spike, protein content and friability. QTLs involved in dormancy and desiccation tolerance were located in *H. vulgare* ssp. *spontaneum* accession Wadi Qilt genotype 23-39 (Zhang *et al*. 2005a). Loci for maximum germination rate under drought stress, and minimum and maximum revival after drought stress were located on chromosome 7H. A new dominant scald resistance gene, *Rrs15* derived from *H. vulgare* ssp. *spontaneum* (accession CPI 77132 Caesarea plant 38), was located on the long arm of chromosome 7H, near the SSR marker HVM49 (Genger *et al*. 2005). In another study using *H. vulgare* ssp. *spontaneum* (accession OUH602), Yun *et al.* (2005) identified a new resistance locus on chromosome 7H for spot blotch (*Rcs2-4*). This gene was located on the short arm of the chromosome in a cluster of genes for resistance to fungal diseases. A third disease resistance study with *H. vulgare* ssp. *spontaneum* (accession ISR42-8) used advanced backcross QTL analysis and located two resistance loci on chromosome 7H, one for powdery mildew (QPm.S42-7H.a) and one for leaf rust (QLr.S42-7H.a), both on the long arm (von Korff *et al*. 2005).

Additional studies located genes and QTLs from cultivated crosses. Emebiri *et al*. (2005b) examined disease resistance in a two-rowed barley population segregating for malting quality traits. The only locus on chromosome 7H, identified by QTL and classical linkage analyses, was for stem rust resistance, likely *Rpg1*. Adult and seedling resistance to spot blotch in Morex was compared in four doubled haploid populations by Bilgic *et al*. (2005). They found that the locus on chromosome 7H, presumably *Rcs5*, was consistently identified for both seedling and adult plant resistance, while loci on other chromosomes were not found in all four populations. Hori *et al*., (2005a) located QTLs for Fusarium head blight resistance from the cultivar Russia 6, along with QTLs for spike morphology. They located QTLs for rachis-internode length and heading date on chromosome 7H.

In a cross between two low protein parents, Emebiri *et al*. (2005a) located QTLs for grain protein content on five chromosomes. The one on chromosome 7H significantly reduced protein in six of the eight environments tested and was not associated with QTLs for yield, height or heading date. Peighambari *et al*. (2005) tested the Steptoe x Morex doubled haploid population for agronomic traits in Iran. Only two QTLs were located on chromosome 7H, for date of spike initiation and 1000 seed weight. Dahleen *et al*. (2005) characterized and located

genes for 27 brachytic semidwarf mutants using SSR markers on near-isogenic lines. One of the new mutants, *brh.v*, was located on chromosome 7H, and the *brh1.z* allele mapped to the expected location of the previously mapped *brh1* locus.

One study has looked at expanding our selection of molecular markers for barley. Zhang *et al*. (2005b) tested 98 EST-SSR markers derived from wheat sequences in barley. They found that 50.4% of the markers amplified sequences in barley. When they examined some of the amplified sequences in more detail, most had repeats similar to those in wheat.

Additional mapping and marker work can be found in proceedings from various meetings, like the North American Barley Researchers Workshop, held in Red Deer Alberta last July.

References:

Bilgic, H., B.J. Steffenson, and P.M. Hayes. 2005. Comprehensive genetic analyses reveal differential expression of spot blotch resistance in four populations of barley. Theor. Appl. Genet. 111:1238-1250.

Dahleen, L.S., L.J. Vander Wal, and J.D. Franckowiak. 2005. Characterization and molecular mapping of genes determining semidwarfism in barley. J. Hered. 96:654-662.

Emebiri, L.C., D.B. Moody, R. Horsley, J. Panozzo, and B.J. Read. 2005a. The genetic control of grain protein content variation in a doubled haploid population derived from a cross between Australian and North American two-rowed barley lines. J. Cereal Sci. 41:107-114.

Emebiri, L.C., G. Platz, and D.B. Moody. 2005b. Disease resistance genes in a doubled haploid population of two-rowed barley segregating for malting quality attributes. Australian J. Agric. Res. 56:49-56.

Genger, R.K., K. Nesbitt, A.H.D. Brown, D.C. Abbott, and J.J. Burdon. 2005. A novel barley scald resistance gene: genetic mapping of the *Rrs15* scald resistance gene derived from wild barley, *Hordeum vulgare* ssp. *spontaneum*. Plant Breeding 124:137-141.

Grando, S., M. Baum, S. Ceccarelli, A. Goodchild, F. Jaby El-Haramein, A. Jahoor, and G. Backes. 2005. QTLs for straw quality characteristics identified in recombinant inbred lines of a *Hordeum vulgare* x *H. spontaneum* cross in a Mediterranean environment. Theor. Appl. Genet. 110:688-695.

Hori, K., T. Kobayashi, K. Sato, and K. Takeda. 2005a. QTL analysis of Fusarium head blight resistance using a high-density linkage map of barley. Theor. Appl. Genet. 111:1661-1672.

Hori, K., K. Sato, N. Nankaku, and K. Takeda. 2005b. QTL analysis in recombinant chromosome substitution lines and doubled haploid lines derived from a cross between *Hordeum vulgare* ssp. *vulgare* and *Hordeum vulgare* ssp. *spontaneum*. Molec. Breeding 16:295-311.

Korff, M. von, H. Wang, J. Léon, and K. Pillen. 2005. AB-QTL analysis in spring barley. 1. Detection of resistance genes against powdery mildew, leaf rust and scald introgressed from wild barley. Theor. Appl. Genet. 111:583-590.

Li, J.Z., X.Q. Huang, F. Heinrichs, M.W. Ganal, and M.S. Röder. 2005. Analysis of QTLs for yield, yield components, and malting quality in a BC_3-DH population of spring barley. Theor. Appl. Genet. 110:356-363.

Peighambari, S.A., B.Y. Samadi, A. Nabipour, G. Charmet, and A. Sarrafi. 2005. QTL analysis for agronomic traits in a barley doubled haploids population grown in Iran. Plant Sci. 169:1008-1013.

Yun, S.J., L. Gyenis, P.M. Hayes, I. Matus, K.P. Smith, B.J. Steffenson, and G.J. Muehlbauer. 2005. Quantitative trait loci for multiple disease resistance in wild barley. Crop Sci. 45:2563-2572.

Zhang, F., G. Chen, Q. Huang, O. Orion, T. Krugman, T. Fahima, A.B. Korol, E. Nevo, and Y. Gutterman. 2005a. Genetic basis of barley caryopsis dormancy and seedling desiccation tolerance at the germination stage. Theor. Appl. Genet. 110:445-453.

Zhang, L.Y., M. Bernard, P. Leroy, C. Feuillet, and P. Sourdille. 2005b. High transferability of bread wheat EST-derived SSRs to other cereals. Theor. Appl. Genet. 111:677-687.

Integrating Molecular and Morphological/Physiological Marker Maps

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Updates to barley morphological/physiological genetic map and gene cloning include publication of the cloning and characterization of the barley *Nec1* locus encoding a cyclic nucleotide-gated ion channel gene (Rostoks *et al.* 2006). The previously reported *rym4* locus coding for the eukaryotic translation initiation factor 4E has been published (Kanyuka *et al.* 2005).

The barley spring vs. winter growth habit candidate genes were cloned and characterized (Von Zitzewitz *et al.* 2005). The *sgh1* locus, renamed *Vrn-H2* to conform with the wheat nomenclature, maps to chromosome 4 (4H) bin 12 approximately 8 cM proximal to *Bmy1* and co-segregating with the *HvSnf2* gene. The *sgh1 (Vrn-H2)* locus, represented by the *ZCCT-H* gene cluster, encodes a dominant transcription factor flowering repressor. Accession numbers of the two closely related candidate genes are AY485977 (*ZCCT-Ha*) and AY485978 (*ZCCT-Hb*). The *Sgh2* locus, renamed *Vrn-H1* to conform to the wheat nomenclature is located on chromosome 7 (5H) bin 11 between markers *Dhn2* and BCD265C (unfortunately designated BCD265B in Zitzewitz *et al*.). However, the gene, designated *HvBM5A*, has been cloned and the sequence is available (AY750995 genomic and AY785826 cDNA cv. Morex sequences, respectively). *HvBM5A* encodes a MADS-box transcription factor. A closely related gene, *HvBM5B*, maps to chromosome 5 (1H) bin 07 closely linked to ABG452. It is proposed that *HvBM5B* represents *Sgh3*, renamed *Vrn-H3* in the wheat nomenclature.

The *Vrs1* gene has been cloned (Komatsuda, Plant & Animal Genome XIV, Abstract W13 p10). Although details have not yet been published, the *Vrs1* locus was reported to encode a homeobox gene.

The cloned morphological/physiological genes represent excellent anchor points for the morphological/physiological barley map since the genes themselves can be used as reference points in mapping populations. There should be ample future opportunities for the identification of other barley genes by homology to the model dicot and monocot plants.

The leaf rust resistance gene *Rph5* was mapped at a high resolution and shown to co-segregate with ABG070 and five ESTs (Mammadov *et al.* 2005). *Rph5* was mapped previously, but this publication provides a high-resolution map and many different closely linked molecular markers.

The location of the non-brittle rachis genes btr1/btr2 was further refined (Senthil and Komatsuda, 2005). However, all of the new markers are AFLP and difficult to integrate with the morphological map.

Molecular mapping located 18 brachytic (*brh*) loci to five of the seven barley chromosomes, albeit with low resolution (Dahleen *et al.* 2005). (The nomenclature of the new loci used here

is that proposed by Dahleen *et al*. for the actual alleles used for mapping see the original paper). The *brh1* and locus was previously mapped with high resolution to chromosome 1 (7H) bin01 and *brh2* was mapped to chromosome 4 (4H) bin 05. Other loci mapping on chromosome 4 (4H) were *brh5* and *brh9*, but lack of flanking markers makes it difficult to determine their bin locations. The same problem exists for *brh3*, *brh4*, and *brh10* loci mapped to chromosome 2 (2H), however *brh3* probably is in bin01. The loci *brh8* and *brh14* were mapped to chromosome 3(3H). A large number of loci were mapped to the short arm of chromosome 7 (5H) including *brh6, brh7, brh11, brh12, brh13, brh17, brh18*. The locus *brh16* was mapped on the long arm of chromosome 1 (7H). Although the sparse markers and lack of flanking markers makes it impossible to reliably place these loci in chromosome bins, they do provide a starting point for those wishing to map these genes more precisely.

The *H. spontaneum* derived leaf scald resistance gene *Rrs15* was mapped to chromosome 1 (7H) long arm 11,5 cM from HVM49 (Genger *et al.* 2005). Since HVM49 is located in bin 12 and the direction of the linkage was not indicated, *Rrs15* could be in bin 11 or 13. The isozyme marker *Acp2* was linked to *Rrs15* at 17.7 cM.

The barley cytoplasmic male sterility restorer gene *Rfm1* was mapped to chromosome 6 (6H) short arm (Murakami *et al.* 2005). Closely linked AFLP markers were identified, however I was not able to assign the locus to a bin.

A very clever use of rice synteny and Arabidopsis was used to identify a cellulose synthase-like (*CslF*) gene cluster as candidates responsible for mediating the cell wall (1,3;1,4)-B-D-glucan syntesis (Burton *et al.* 2006). The work was initiated from the map location of a major QTL for (1,3;1,4)-B-D-glucan content of un-germinated barley grains on chromosome 2 (2H). This QTL is located between the markers *Adh8* bin 6 and ABG019 bin 7 with the peak closer to ABG019. Therefore, I have assigned the *CslF* locus to 2 (2H) bin 7. I believe this is the first example of a map-based cloning of a QTL in barley.

Please advise me of any additions or corrections to this information.

Bin Assignments for Morphological Map Markers and closest molecular marker

Chr.1 (7H)			
BIN1	ABG704		
	*Rpg1	RSB228	Brueggeman *et al*., PNAS 99:9328, ‘02
		*Run1	
	Rdg2a	MWG851A	Bulgarelli *et al*., TAG 108:1401, ‘04
	Rrs2	MWG555A	Schweizer *et al*., TAG 90:920, ‘95
		mlt	
	brh1	MWG2074B	Li et al., 8th IBGS 3:72, ‘00
BIN2	ABG320		
	Est5	iEst5	Kleinhofs *et al*., TAG 86:705, ‘93
	fch12	BCD130	Schmierer *et al*., BGN 31:12, ‘01
	*wax	Wax	Kleinhofs BGN 32:152, ‘02
	gsh3	His3A	Kleinhofs BGN 32:152, ‘02

BIN3	ABC151A		
	fch5	ABC167A	Kleinhofs BGN 32:152, ‘02
	Rcs5	KAJ185	Johnson & Kleinhofs, unpublished
	yvs2		
	cer-ze	ABG380	Kleinhofs BGN 27:105, ‘96
BIN4	ABG380		
	wnd		
	Lga	BE193581	Johnson & Kleinhofs, unpublished
	abo7		
BIN5	ksuA1A		
	ant1		
	nar3	MWG836	Kleinhofs BGN 32:152, ‘02
	ert-m		
	ert-a		
BIN6	ABC255		
	ert-d		
	fch8		
	fst3		
	cer-f		
	msg14		
BIN7	ABG701		
	dsp1	cMWG704	Sameri (in press)
	msg10		
	rsm1	ABC455	Edwards & Steffenson, Phytopath. 86:184,’96
	sex6		
	seg5		
	seg2		
	pmr	ABC308	Kleinhofs BGN 27:105, ‘96
	mo6b	Hsp17	Soule *et al*., J Her. 91:483, ‘00
	nud	CDO673	Heun *et al*., Genome 34:437, ‘91
	fch4	MWG003	Kleinhofs BGN 27:105, ‘96
BIN8	*Amy2	Amy2	Kleinhofs *et al*., TAG 86:705, ‘93
	lks2	WG380B	Costa *et al*., TAG 103:415, ‘01
	Rpt4	Psr117D	Williams *et al*., TAG 99:323, ‘99
	ubs4		
	blx2		
BIN9	RZ242		
	lbi3		
	xnt4		
	lpa2	?	Larson *et al*., TAG 97:141, ‘98
	msg50		
	Rym2		
	seg4		
BIN10	ABC310B		
	Xnt1	BF626025	Hansson et *al*., PNAS 96:1744, ‘99
	xnt-h	BF626025	Hansson et *al*., PNAS 96:1744, ‘99
BIN11	ABC305		
	Rph3		
	Tha2		Toojinda *et al*., TAG 101:580, ‘00
BIN12	ABG461A		

Mlf
xnt9
seg1
msg23
BIN13 Tha
Rph19 Rlch4(Nc) Park & Karakousis Plt. Breed. 121:232. '02

Chr.2 (2H)
BIN1 MWG844A
sbk
brh3 Bmac0134 Dahleen *et al.*, J. Heredity 96:654, '05
BIN2 ABG703B
BIN3 MWG878A gsh6 Kleinhofs BGN 32:152, '02
gsh1
gsh8
BIN4 ABG318
Eam1
Ppd-H1 MWG858 Laurie *et al.*, Heredity 72:619, '94
sld2
rtt
flo-c
sld4
BIN5 ABG358
fch15
brc1
com2
BIN6 Pox
msg9
abo2
Rph15 P13M40 Weerasena *et al.*, TAG 108:712 '04
rph16 MWG874 Drescher *et al.*, 8thIBGS II:95, '00
BIN7 Bgq60
yst4 CDO537 Kleinhofs BGN 32:152, '02
Az94 CDO537 Kleinhofs BGN 32:152, '02
gai MWG2058 Börner *et al.*, TAG 99:670, '99
msg33
*HvCslF (barley Cellulose synthase-like) Burton *et al.*, Science 311:1940 '06
*Bmy2
msg3
fch1
BIN8 ABC468
Eam6 ABC167b Tohno-oka et al., 8thIBGS III:239, '00
gsh5
msg2
eog ABC451 Kleinhofs BGN 27:105, '96
abr
cer-n
BIN9 ABC451
Gth

	hcm1		
	wst4		
	*vrs1	MWG699	Komatsuda *et al*., Genome 42:248, ‘00
BIN10	MWG865		
	cer-g		
	Lks1		
	mtt4		
	Pre2		
	msg27		
BIN11	MWG503		
	Rha2	AWBMA21	Kretschmer *et al*., TAG 94:1060, ‘97
	Ant2	MWG087	Freialdenhoven *et al*., Plt. Cell 6:983, ’94
	*Rar1	AW983293B	Freialdenhoven et al., Plt. Cell 6:983, ’94
	fol-a		
	gal	MWG581A	Börner *et al*., TAG 99:670, ‘99
	fch14		
	Pau		
BIN12	ksuD22		
	Pvc		
BIN13	ABC252		
	lig	BCD266	Pratchett & Laurie Hereditas 120:35, ‘94
	nar4	Gln2	Kleinhofs BGN 27:105, ‘96
	Zeo1	cnx1	Costa *et al*., TAG 103:415, ‘01
	lpa1	ABC157	Larson *et al*., TAG 97:141, ‘98
BIN14	ABC165		
BIN15	MWG844B		
	gpa	CDO036	Kleinhofs BGN 27:105, ‘96
	wst7	MWG949A	Costa *et al*., TAG 103:415, ‘01
	MlLa	Ris16	Giese *et al*., TAG 85:897, ‘93
	trp		

Chr. 3 (3H)

BIN1	Rph5	ABG070	Mammadov *et al*., TAG 111:1651, ‘05
	Rph6	BCD907	Zhong *et al*., Phytopath. 93:604, ‘03
	Rph7	MWG848	Brunner *et al*., TAG 101:783, ‘00
BIN2	JS195F	BI958652; BF631357; BG369659	
	ant17		
	sld5		
	mo7a	ABC171A	Soule *et al*., J. Hered. 91:483, ‘00
	brh8		
BIN3	ABG321		
	xnt6		
BIN4	MWG798B		

	btr1		Senthil & Komatsuda Euphytica 145:215, ‘05
	btr2		Senthil & Komatsuda Euphytica 145:215, ‘05
	lzd		
	alm	ABG471	Kleinhofs BGN 27:105, ‘96
BIN5	BCD1532		
	abo9		
	sca		
	yst2		
	dsp10		
BIN6	ABG396		
	Rrs1		Graner *et al*., TAG 93: 421 ´96
	Rh/Pt	ABG396	Smilde *et al*., 8th IBGS 2:178, ‘00
	Rrs.B87	BCD828	Williams *et al*., Plant Breed. 120:301, ‘01
	AtpbB		
	abo6		
	xnt3		
	msg5		
	ari-a		
	yst1		
	zeb1		
	ert-c		
	ert-ii		
	cer-zd		
	Ryd2	WG889B	Collins *et al*., TAG 92:858, ‘96
	*uzu	AB088206	Saisho *et al*., Breeding Sci. 54:409, ‘04
BIN7	MWG571B		
	cer-r		
BIN8	ABG377		
	wst6		
	cer-zn		
	sld1		
BIN9	ABG453		
	wst1		
BIN10	CDO345		
	vrs4		
	Int1		
	gsh2		
BIN11	CDO113B		
	als		
	sdw1	PSR170	Laurie *et al*., Plant Breed. 111:198, ‘93
BIN12	His4B		
	sdw2		
BIN13	ABG004		
	Pub	ABG389	Kleinhofs *et al*., TAG 86:705, ‘93
BIN14	ABC161		
	cur2		
BIN15	ABC174		
	Rph10		
	fch2		
BIN16	ABC166		

	eam10		
	Est1/2/3		
	*rym4	*eIF4E*	Stein *et al.*,Plt. J. 42:912, ‘05
	*rym5	*eIF4E*and	Kanyuka *et al.*, Mol. Plant Path. 6:449, ’05
	Est4		
	ant28		

Chr.4 (4H)

BIN1	MWG634		
BIN2	JS103.3		
	fch9		
	sln		
BIN3	Ole1	Dwf2	Ivandic *et al.*, TAG 98:728, ‘99
	Ynd		
	int-c	MWG2033	Komatsuda, TAG 105:85, ‘02
	Zeo3		
	glo-a		
	rym1	MWG2134	Okada *et al.*, Breeding Sci. 54:319, ‘04
BIN4	BCD402B		
	*Kap	X83518	Müller *et al.*, Nature 374:727, ‘95
	lbi2		
	zeb2		
	lgn3		
BIN5	BCD808B		
	lgn4		
	lks5		
	eam9		
	msg24		
BIN6	ABG484		
	glf1		
	rym11	MWG2134	Bauer *et al.*, TAG 95:1263, ‘97
	Mlg	MWG032	Kurth *et al.*, TAG 102:53, ‘01
	cer-zg		
	brh2		
BIN7	bBE54A		
	glf3		
	frp		
	min1		
	blx4		
	sid		
	blx3		
BIN8	BCD453B		
	blx1		
BIN9	ABG319A		
	ert1		
BIN10	KFP221		
	*mlo	P93766	Bueschges *et al.*, Cell 88:695, ‘97
BIN11	ABG397		
BIN12	ABG319C		

	Hsh	HVM067	Costa *et al*., TAG 103:415, ‘01
	Hln		
	*sgh1	(ZCCT-H; HvSnf2)	Zitzewitz *et al*., PMB 59:449, ‘05
	yhd1		
BIN13	*Bmy1	pcbC51	Kleinhofs *et al*., TAG 86:705, ‘93
	rym8	MWG2307	Bauer *et al*., TAG 95:1263, ‘97
	rym9	MWG517	Bauer *et al*., TAG 95:1263, ‘97
	Wsp3		

Chr. 5 (1H)

BIN1	Tel5P		
	Rph4		
	Mlra		
	Cer-yy		
	Sex76	Hor2	Netsvetaev BGN 27:51, ‘97
	Hor5	Hor5	Kleinhofs *et al*., TAG 86:705, ‘93
BIN2	MWG938		
	*Hor2	Hor2	Kleinhofs *et al*., TAG 86:705, ‘93
	Rrs14	Hor2	Garvin *et al*., Plant Breed. 119:193-196, ‘00
	*Mla6	AJ302292	Halterman *et al*., Plt J. 25:335, ‘01
BIN3	MWG837		
	*Hor1	Hor1	Kleinhofs *et al*., TAG 86:705, ‘93
	Rps4		
	Mlk		
BIN4	ABA004		
	Lys4		
BIN5	BCD098		
	Mlnn; msg31; sls; msg4; fch3;		
BIN6	Ica1		
	amo1		
BIN7	JS074		
	clh		
	vrs3		
	Ror1	ABG452	Collins *et al*., Plt. Phys. 125:1236, ‘01
	*Sgh3	(HvBM5B)	Zitzewitz *et al*., PMB 59:449, ‘05
BIN8	Pcr2		
	fst2		
	cer-zi		
	cer-e		
	ert-b		
	MlGa		
	msg1		
	xnt7		
BIN9	Glb1		
	*nec1	BF630384	Rostoks *et al*., MGG 275:159, ‘06
BIN10	DAK123B		
	abo1		
	Glb1		
BIN11	PSR330		

	wst5		
	cud2		
BIN12	MWG706A		
	rlv		
	lel1		
BIN13	BCD1930		
	Blp	ABC261	Costa *et al*., TAG 103:415, ‘01
BIN14	ABC261		
	fch7		
	trd		
	eam8		

Chr. 6 (6H)

BIN1	ABG062		
	*Nar1	X57845	Kleinhofs *et al*., TAG 86:705, ‘93
	abo15		
BIN2	ABG378B		
	nar8	ABG378B	Kleinhofs BGN 27:105, ‘96
	nec3		
	Rrs13		
BIN3	MWG652A		
BIN4	DD1.1C		
	msg36		
BIN5	ABG387B		
	nec2		
	ant21		
	msg6		
	eam7		
BIN6	Ldh1		
	rob	HVM031	Costa *et al*., TAG 103:415, ‘01
	sex1		
	gsh4		
	ant13		
	cul2	Crg4(KFP128)	Babb & Muehlbauer BGN 31:28, ‘01
	fch11		
	mtt5		
	abo14		
BIN7	ABG474		
BIN8	ABC170B		
BIN9	*Nar7	X60173	Warner *et al*., Genome 38:743, ‘95
	*Amy1	JR115	Kleinhofs *et al*., TAG 86:705, ‘93
	*Nir	pCIB808	Kleinhofs *et al*., TAG 86:705, ‘93
	mul2		
	cur3		
BIN10	MWG934		
	lax-b		
	raw5		
	cur1		
BIN11	Tef1		

BIN12	xnt5		
	Aat2		
BIN13	Rph11	Acp3	Feuerstein *et al.*, Plant breed. 104:318, '90
	lax-c		
BIN14	DAK213C		
	dsp9		

Chr. 7 (5H)

BIN1	DAK133		
	abo12		
	msg16		
	ddt		
BIN2	MWG920.1A		
	dex1		
	msg19		
	nld		
	fch6		
	glo-b		
BIN3	cud1	ABG705A	
	lys3		
	fst1		
	blf1		
	vrs2		
BIN4	ABG395		
	cer-zj		
	cer-zp		
	msg18		
	wst2		
	Rph2	ITS1	Borovkova *et al.*, Genome 40:236, '97
	lax-a	PSR118	Laurie *et al.*, TAG 93:81, '96
	com1		
	ari-e		
	ert-g		
	ert-n		
BIN5	Ltp1		
	rym3	MWG028	Saeki *et al.*, TAG 99:727, '99
BIN6	WG530		
BIN7	ABC324		
BIN8	ABC302A		
BIN9	BCD926		
	srh	ksuA1B	Kleinhofs *et al.*, TAG 86:705, '93
	cer-i		
	mtt2		
	lys1		
	cer-t		
	dsk		
	var1		
	cer-w		

	Eam5		
BIN10	ABG473		
	raw1		
	msg7		
BIN11	MWG514B		
	Rph9/12	ABG712	Borokova *et al*., Phytopath. 88:76, ‘98
	*Sgh2 (HvBM5A)		Zitzewitz *et al*., PMB 59:449, ‘05
	*Ror2	AY246906	Collins *et al*., Nature 425:973, ‘03
	lbi1		
	Rha4		
	raw2		
BIN12	WG908		
	none		
BIN13	ABG496		
	rpg4	ARD5303	Druka *et al*., unpublished
	RpgQ	ARD5304	Druka *et al*., unpublished
BIN14	ABG390		
	var3		
BIN15	ABG463		

BIN markers are indicated
* - indicates the gene has been cloned

References:

Babb, S.L. and G.J. Muehlbauer. 2001. Map location of the Barley Tillering Mutant *uniculm2* (*cul2*) on Chromosome 6H. BGN31:28.

Bauer, E., J. Weyen, A. Schiemann, A. Graner, and F. Ordon. 1997. Molecular mapping of novel resistance genes against Barley Mild Mosaic Virus (BaMMV). Theor. Appl. Genet. 95:1263-1269.

Borovkova, I.G., Y. Jin, B.J. Steffenson, A. Kilian, T.K. Blake, and A. Kleinhofs. 1997. Identification and mapping of a leaf rust resistance gene in barley line Q21861. Genome 40:236-241.

Borokova, I.G., Y. Jin, and B.J. Steffenson. 1998. Chromosomal Location and Genetic Relationship of Leaf Rust Resistance Genes *Rph9* and *Rph12* in Barley. Phytopathology 88:76-80.

Börner, A., V. Korzun, S. Malyshev, V. Ivandic, and A. Graner. 1999. Molecular mapping of two dwarfing genes differing in their GA response on chromosome 2H of barley. Theor. Appl. Genet. 99:670-675.

Brueggeman, R., N. Rostoks, D. Kudrna, A. Kilian, F. Han, J. Chen, A. Druka, B. Steffenson, and A. Kleinhofs. 2002. The barley stem-rust resistance gene *Rpg1* is a novel disease-resistance gene with homology to receptor kinases. Proc. Natl. Acad. Sci. USA 99:9328-9333.

Brunner, S., B. Keller, and C. Feuillet. 2000. Molecular mapping of the Rph7.g leaf rust resistance gene in barley (*Hordeum vulgare* L.). Theor. Appl. Genet. 101:763-788.

Büschges, R., K. Hollricher, R. Panstruga, G. Simons, M. Wolter, A. Frijters, R. van Daelen, T. van der Lee, P. Diergaarde, J. Groenendijk, S. Töpsch, P. Vos, F. Salamini, and P. Schulze-Lefert. 1997. The barley *mlo* gene: A novel control element of plant pathogen resistance. Cell 88:695-705.

Bulgarelli, D., N.C. Collins, G. Tacconi, E. Dellaglio, R. Brueggeman, A. Kleinhofs, A.M. Stanca, and G. Vale. 2004. High resolution genetic mapping of the leaf stripe resistance gene Rdg2a in barley. Theor. Appl. Genet. 108:1401-1408.

Burton, R.A., S.M. Wilson, M. Hrmova, A.J. Harvey, N.J. Shirley, A. Medhurst, B.A. Stone, E.J. Newbigin, A. Bacic, and G.B. Fincher. 2006. Cellulose synthase-like CslF genes mediate the synthesis of cell wall (1,3; 1,4)-b-D-glucans. Science 311:1940-1943.

Collins, N.C., N.G. Paltridge, C.M. Ford, and R.H. Symons.1996. The *Yd2* gene for barley yellow dwarf virus resistance maps close to the centromere on the long arm of barley chromosome 3. Theor. Appl. Genet. 92:858-864.

Collins, N.C., T. Lahaye, C. Peterhänsel, A. Freialdenhoven, M. Corbitt, and P. Schulze-Lefert. 2001. Sequence haplotypes revealed by sequence-tagged site fine mapping of

the *Ror1* gene in the centromeric region of barley chromosome 1H. Plant Physiology 125:1236-1247.

Collins, N.C., H. Thordal-Christensen, V. Lipka, S. Bau, E. Kombrink, J-L. Qiu, R. Hückelhoven, N. Stein, A. Freialdenhoven, S.C. Somerville, and P. Schulze-Lefert. 2003. Snare-protein-mediated disease resistance at the plant cell wall. Nature 425:973-976.

Costa, J.M., A. Corey, M. Hayes, C. Jobet, A. Kleinhofs, A. Kopisch-Obusch, S.F. Kramer, D. Kudrna, M. Li, O. Piera-Lizaragu, K. Sato, P. Szues, T. Toojinda, M.I. Vales, and R.I. Wolfe. 2001. Molecular mapping of the Oregon Wolfe Barleys: a phenotypically polymorphic doubled-haploid population. Theor. Appl. Genet. 103:415-424.

Drescher, A., V. Ivandic, U. Walther, and A. Graner. 2000. High-resolution mapping of the Rph16 locus in barley. p. 95-97. *In*: S. Logue (ed.) Barley Genetics VIII. Volume II. Proc. Eigth Int. Barley Genet. Symp. Adelaide. Dept. Plant Science, Waite Campus, Adelaide University, Glen Osmond, South Australia.

Dahleen, L.S., L.J. Vander Wal, and J.D. Franckowiak. 2005. Characterization and molecular mapping of genes determining semidwarfism in barley. J. Heredity 96:654-662.

Edwards, M.C. and B.J. Steffenson. 1996. Genetics and mapping of barley stripe mosaic virus resistance in barley. Phytopath. 86;184-187.

Feuerstein, U., A.H.D. Brown, and J.J. Burdon. 1990. Linkage of rust resistance genes from wild barley (*Hordeum spontaneum*) with isoenzyme markers. Plant. Breed. 104:318-324.

Freialdenhoven, A., B. Scherag, K. Hollrichter, D.B. Collinge, H. Thordal-Christensen, and P. Schulze-Lefert. 1994. *Nar-1* and *Nar-2*, two loci required for *Mla12*-specified race-specific resistance to powdery mildew in barley. Plant Cell 6:983-994.

Garvin, D.F., A.H.D. Brown, H. Raman, and B.J. Read. 2000. Genetic mapping of the barley *Rrs14* scald resistance gene with RLFP, isozyme and seed storage protein marker. Plant Breeding 119:193-196.

Genger, R.K., K. Nesbitt, A.H.D. Brown, D.C. Abbott, and J.J. Burdon. 2005. A novel scald resistance gene: genetic mapping of the Rrs15 scald resistance derived from wild barley, Hordeum vulgare ssp. spontaneum. Plant Breeding 124:137-141.

Giese, H., A.G. Holm-Jensen, H.P. Jensen, and J.Jensen. 1993. Localisation of the Laevigatum powdery mildew resistance gene to barley chromosome 2 by the use of RLFP markers. Theor. Appl. Genet. 85:897-900.

Graner, A. and A. Tekauz. 1996. RFLP mapping in barley of a dominant gene conferring to scald (*Rynchosporium secalis*). Theor. Appl. Genet. 93:421-425.

Haltermann, D., F. Zhou, F. Wei, R.P. Wise, and P. Schulze-Lefert. 2001. The MLA6 coiled coil, NBS-LRR protein confers *AvrMla6*-dependent resistance specificity to *Blumeria graminis* f. sp. *hordei* in barley and wheat. Plt. J. 25:335-348.

Hansson, A., C.G. Kannangara, D. von Wettstein, and M. Hansson. 1999. Molecular basis for semidomiance of missense mutations in the XANTHA-H (42-kDa) subunit of magnesium chelatase. Proc. Natl. Acad. Sci. USA 96:1744-1749.

Heun, M., A.E. Kennedy, J.A. Anderson, N.L.V. Lapitan, M.E. Sorrells, and S.D. Tanksley. 1991. Construction of a restriction fragment length polymorphism map for barley (*Hordeum vulgare*). Genome 34:437-447.

Ivandic, V., S. Malyshev, V. Korzun, A. Graner, and A. Börner. 1999. Comparative mapping of a gibberellic acid-insensitive dwarfing gene (*Dwf2*) on chromosome 4HS in barley. Theor. Appl. Genet. 98:728-731.

Kanyuka, K., A. Druka, D..G. Caldwell, A. Tymon, N. McCallum, R. Waugh, and M. J. Adams. 2005. Evidence that the recessive bymovirus resistance locus *rym4* in barley corresponds to the eukaryotic translation initiation factor 4E gene. Molecular Plant pathology 6:449-458.

Kleinhofs, A., A. Kilian, M.A. Saghai Marrof, R.M. Biyashev, P. Hayes, F.Q. Chen, N. Lapitan, A. Fenwick, T.K. Blake, V. Kanazin, E. Ananiev, L. Dahleen, D. Kudrna, J. Bollinger, S.J. Knapp, B. Liu, M. Sorells, M. Heun, J.D. Franckowiak, D. Hoffman, R. Skadsen, and B.J. Steffenson. 1993. A molecular, isozyme and morphologicsl map of the barley (*Hordeum vulgare*) genome. Theor. Appl. Genet. 86:705-712.

Kleinhofs, A. 1996. Integrating Barley RFLP and Classical Marker Maps. Coordinator's report. BGN27:105-112.

Kleinhofs, A. 2002. Integrating Molecular and Morphological/Physiological Marker Maps. Coordinator's Report. BGN32:152-159.

Komatsuda, T., W. Li, F. Takaiwa, and S. Oka. 1999. High resolution map around the *vrs1* locus controlling two- and six-rowed spike in barley. (*Hordeum vulgare*). Genome 42:248-253.

Komatsuda, T., and Y. Mano. 2002. Molecular mapping of the intermedium spike-c (*int-c*) and non-brittle rachis 1 (*btr1*) loci in barley (*Hordeum vulgare* L.). Theor. Appl Genet. 105:85-90.

Komatsuda, T., M. Pourkheirandish, C. He, P. Azhaguvel, H. Kanamori, D. Perovic, N. Stein, A. Graner, U. Lundqvist, T. Fujimura, M. Matsuoka, T. Matsumoto, and M. Yano. 2006. Map-based cloning of the barley six-rowed spike gene *vrs1*. Plant and Animal Genome XIV. San Diego, USA, Abstract W13, p.10.

Kretschmer, J.M., K.J. Chalmers, S. Manning, A. Karakousis, A.R. Barr, A.K.M.R. Islam, S.J. Logue, Y.W. Choe, S.J. Barker, R.C.M. Lance, and P. Langridge. 1997. RFLP mapping of the *Ha2* cereal cyst nematode resistance in barley.Theor. Appl. Genet. 94:1060-1064.

Kurth, J., R. Kolsch, V. Simons, and P. Schulze-Lefert. 2001. A high-resolution genetic map and a diagnostic RFLP marker for the *Mlg* resistance locus to powdery mildew in barley. Theor. Appl. Genet. 102:53-60.

Larson, S.R., K.A. Young, A. Cook, T.K. Blake, and V. Raboy. 1998. Linkage mapping of two mutations that reduce phytic acid content of barley grain. Theor. Appl. Genet. 97:141-146.

Laurie, D.A., N. Pratchett, C. Romero, E. Simpson, and J.W. Snape. 1993. Assignment of the *denso* dwarfing gene to the long arm of chromosome 3 (3H) of barley by use of RFLP markers. Plant. Breed. 111:198-203.

Laurie, D.A., N. Pratchett, J.H. Bezan, and J.W. Snape. 1994. Genetic analysis of a photoperiod response gene on the short arm of chromosome 2 (2H) of Hordeum vulgare (barley). Heredity 72:619-627.

Laurie, D.A., N. Pratchett, R.A. Allen, and S.S. Hantke. 1996. RFLP mapping of the barley homeotic mutant *lax-a*. Theor. Appl. Genet. 93:81-85.

Li. M., D. Kudrna, and A. Kleinhofs. 2000. Fine mapping of a Semi-dwarf gene Brachytic1 in barley. p. 72-74. *In*: S. Logue (ed.) Barley Genetics VIII. Volume III. Proc. Eigth Int. Barley Genet. Symp. Adelaide. Dept. Plant Science, Waite Campus, Adelaide University, Glen Osmond, South Australia.

Mammadov, J.A., B. J. Steffenson, and M.A. Saghai Maroof. 2005. High resolution mapping of the barley leaf rust resistance gene Rph5 using barley expressed sequence tags (ESTs) and synteny with rice. Theor. Appl. Genet. 111:1651-1660.

Murakami, S., K. Matsui, T. Komatsuda, and Y. Furuta. 2005. AFLP-based STS markers closely linked to a fertility restoration locus (Rfm1) for cytoplasmic male sterility in barley. Plant Breeding 124:133-136.

Müller, K.J., N. Romano, O. Gerstner, F. Gracia-Maroto, C. Pozzi, F. Salamini, and W. Rhode. 1995. The barley *Hooded* mutation caused by a duplication in a homeobox gene intron. Nature 374:727-730.

Netsvetaev, V.P. 1997. High lysine mutant of winter barley - L76. BGN27:51-54.

Okada, Y., R. Kanatani, S. Arai, and I Kazutoshi. 2004. Interaction between barley mosaic disease-resistance genes *rym1* and *rym5*, in the response to BaYMV strains. Breeding Science 54 (4):319-324.

Park, R.F. and A. Karakousis. 2002. Characterization and mapping *Rph19* conferring resistance to *Puccinia hordei* in the cultivar 'Rea1' and several Australian barley. Plant Breeding 121:232-236.

Pratchett, N. and D.A. Laurie. 1994. Genetic map location of the barley developmental mutant *liguleless* in relation to RFLP markers. Hereditas 120:35-39.

Rostoks, N., D. Schmierer, S. Mudie, T. Drader, R. Brueggeman, D. Caldwell, R. Waugh, and A. Kleinhofs. 2006. Barley necrotic locus *nec1* encodes the cyclic nucleotide-gated ion channel 4 homologous to the *Arabidopsis Hlm1*. Mol.Gen. Genomics 275:159-168.

Saeki, K., C. Miyazaki, N. Hirota, A. Saito, K. Ito, and T. Konishi. 1999. RFLP mapping of BaYMV resistance gene *rym3* in barley (*Hordeum vulgare*). Theor. Appl. Genet. 99:727-732.

Saisho, D., K.-I. Tanno, M. Chono, I. Honda, H. Kitano, and K. Takeda. 2004. Spontaneous Brassinolide-insensitive barley mutants 'uzu' adapted to East Asia. Breeding Science 54(4): 409-416.

Schmierer, D., A. Druka, D. Kudrna, and A. Kleinhofs. 2001. Fine Mapping of the *fch12* chlorina seedling mutant. BGN31:12-13.

Schweizer, G.F., M. Baumer, G. Daniel, H. Rugel, and M.S. Röder. 1995. RFLP markers linked to scald (*Rhynchosporium secalis*) resistance gene *Rh2* in barley. Theor. Appl. Genet. 90:920-922.

Smilde, W.D., A. Tekauz, and A. Graner. 2000. Development of a high resolution map for the *Rh* and *Pt* resistance on barley Chromosome 3H. p. 178-180. *In*: S. Logue (ed.) Barley Genetics VIII. Volume II. Proc. Eigth Int. Barley Genet. Symp. Adelaide. Dept. Plant Science, Waite Campus, Adelaide University, Glen Osmond, South Australia.

Soule, J.D., D.A. Kudrna, and A. Kleinhofs. 2000. Isolation, mapping, and characterization of two barley multiovary mutants. J. Heredity 91:483-487.

Senthil, N. and T. Komatsuda. 2005. Inter-subspecific maps of non-brittle rachis gene btr1/btr2 using occidental, oriental and wild barley lines. Euphytica 145:215-220.

Stein, N., D. Perovic, J. Kumlehn, B. Pellio, S. Stracke, S. Streng, F. Ordon, and A. Graner. 2005. The eukaryotic translation initian factor 4E confers multiallelic recessive Bymovirus resistance in *Hordeum vulgare* (L.). The Plant Journal 42:912-922.

Tohno-oka, T., M. Ishit, R. Kanatani, H. Takahashi, and K. Takeda. 2000. Genetic Analysis of photoperiotic response of barley in different daylength conditions. p.239-241. *In*: S. Logue (ed.) Barley Genetics VIII. Volume III. Proc. Eigth Int. Barley Genet. Symp. Adelaide. Dept. Plant Science, Waite Campus, Adelaide University, Glen Osmond, South Australia.

Toojinda, T., L.H. Broers, X.M. Chen, P.M. Hayes, A. Kleinhofs, J. Korte, D. Kudrna, H. Leung, R.F. Line, W. Powell, L. Ramsey, H. Vivar, and R. Waugh. 2000. Mapping quantitative and qualitative disease resistance genes in a doubled haploid population of barley (*Hordeum vulgare*). Theor. Appl. Genet. 101:580-589.

Warner, R.L., D.A. Kudrna, and A. Kleinhofs. 1995. Association of the NAD(P)H-bispecific nitrate reductase structural gene with the *Nar7* locus in barley. Genome 38:743-746.

Weerasena, J.S., B.J. Steffenson, and A.B. Falk. 2004. Conversion of an amplified fragment length polymorphism marker into a c-dominant marker in mapping *Rph15* gene

conferring resistance to barley leaf rust, *Puccinia hordei* Otth. Theor. Appl. Genet. 108:712-719.

Williams, K.J., A. Lichon, P. Gianquitto, J.M. Kretschmer, A. Karakousis, S. Manning, P. Langridge, and H. Wallwork. 1999. Identification and mapping of a gene conferring resistance to the spot form of net blotch (*Pyrenophora teres* f. *maculata*) in barley. Theor. Appl. Genet. 99: 323-327.

Williams, K., P. Bogacki, L. Scott, A. Karakousis, and H. Wallwork. 2001. Mapping of a gene for leaf scald resistance in barley line 'B87/14' and validation of microsatellite and RFLP markers for marker-assisted selection. Plant Breed. 120:301-304.

Zitzewitz, J. von, P. Scucs, J. Dobcov, L. Yan, E. Francia, N. Pecchioni, A. Casas, T.H.H. Chen, P.M. Hayes, and J.S. Skinner. 2005. Molecular and structural characterization of barley vernalization genes. Plant Molecular Biology 59:449-467.

Zhong, S.B., R.J. Effertz, Y. Jin, J.D. Franckowiak, and B.J. Steffenson. 2003. Molecular mapping of the leaf rust resistance gene *Rph6* in barley and its linkage relationships with *Rph5* and *Rph7*. Phytopathology 93 (5):604-609.

Coordinator's report: Barley Genetic Stock Collection

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In 2005, 373 barley genetic stocks were planted in the field and in the greenhouse for evaluation and for seed increase.

Four mapping populations including SSD F6 OSU 1/Harr, SSD F6 OSU 2/Harr, SSD F6 OSU 11/Harr and SSD F6 OSU 15/Harr derived from crosses between *Hordeum vulgare subps. Spontaneum* with the cultivar "Harrington" and 142 *H. spontaneum* introgression lines BC2 S1 and BC2 S5 were obtained from Dr. Pat Hayes, OSU and maintained at Aberdeen. Two populations, SSD F6 OSU 1/Harr and SSD F6 2/Harr were planted in the field for seed increase.

One hundred thirty-two samples of barley genetic stocks were shipped to researchers in 2005.

Coordinator's report: Trisomic and aneuploid stocks

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There is no new information about trisomic and aneuploid stocks. A list of these stocks are available in BGN 25:104. Seed request for this stock should be sent to the coordinator.

Coordinator's report: Autotetraploids

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The collection of barley autotetraploids (exclusively spring types) described in former issues of BGN is maintained at the Giessen Field Experiment Station of our institute. The set of stocks, i.e. autotetraploids (4n) and corresponding diploid (2n) progenitors (if available) have last been grown in the field for seed multiplication in summer 2000. Limited seed samples of the stocks are available for distribution.

Coordinator's report: The Genetic Male Sterile Barley Collection

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The GMSBC has been at Brandon since 1992. If there are any new sources of male-sterile genes that you are aware of, please advice me, as this would be a good time to add any new source to the collection. For a list of the entries in the collection, simply E-mail me at the above adress. I can send the file (14Mb) in Excel format. We continue to store the collection at -20^{o}C and will have small (5 g) samples available for the asking. Since I have not received any reports or requests the last years, there is absolutely no summary in my report.

Coordinator's report: Translocations and balanced tertiary trisomics

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Chromosome 5H of *Hordeum vulgare* carries a gene(s) that accelerates heading in a wheat background. To introduce the early heading gene(s) of barley into the wheat genome, the Japanese scientists S. Taketa and colleagues attempted to induce homoeologous recombination between wheat and 5H chromosomes by 5B nullisomy. A nullisomic 5B, trisomic 5A, monosomic 5H plant (2n = 42) was produced from systematic crosses between aneuploid stocks of wheat group 5 chromosomes. Twelve plants (1.8%) were selected as putative wheat-barley 5H recombinants. Cytological analyses using fluorescence in situ hybridization and C-banding revealed that 6 of the progeny lines are true homoeologous recombinants between the long arms of chromosomes 5D and 5H. The 6 cytologically confirmed recombinant lines included only 2 types (3 lines each), which were reciprocal products derived from exchanges at the same distal interval defined by two flanking markers. One type had a small 5HL segment translocated to the 5DL terminal, and the other type had a small terminal 5DL segment translocated to the 5HL terminal. In the latter type, the physical length of translocated barley segments slightly differed among lines.

There were no requests for samples of balanced tertiary trisomics or tranlocation lines. The collection is being maintained in cold storage. To the best knowledge of the coordinator, there are no new publications dealing with balanced tertiary trisomics in barley. Limited seed samples are available any time, and requests can be made to the coordinator.

Reference:

Taketa, S, T. Awayama , M. Ichii, M. Sunakawa, T. Kawahara, and K. Murai. 2005. Molecular cytogenetic identification of nullisomy 5B induced homoeologous recombination between wheat chromosome 5D and barley chromosome 5H. Genome 48: 115-124.

Coordinator's report: *Eceriferum* Genes

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No research work on gene localization has been reported on the collections of *Eceriferum* and *Glossy* genes since the latest reports in Barley Genetics Newsletter (BGN). All information and descriptions done in Barley Genetics Newsletter (BGN) Volume 26 are valid and still up-to-date. The database of the Swedish collection has been updated during the last months and will soon be searchable within International European databases. All Swedish *Eceriferum* alleles can be seen in the SESTO database of the Nordic Gene Bank. As my possibilities in searching literature are very limited, I apologize if I am missing any important papers. Please send me notes of publications and reports to include in next year's reports. Descriptions, images and graphic chromosome map displays of the *Eceriferum* and *Glossy* genes are available in the AceDB database for Barley Genes and Barley Genetic Stocks, and they get currently updated. Its address is found by: www.untamo.net/bgs

Every research of interest in the field of *Eceriferum* genes, 'Glossy sheath' and 'Glossy leaf' genes can be reported to the coordinator as well. Seed requests regarding the Swedish mutants can be forwarded to the coordinator udda@ngb.se or udda@nordgen.org or to the Nordic Gene Bank, www.nordgen.org/ngb, all others to the Small Grain Germplasm Research Facility (USDA-ARS), Aberdeen, ID 83210, USA, anhang@uida.edu or to the coordinator at any time.

Coordinator's report: Nuclear genes affecting the chloroplast

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Chlorophyll biosynthesis is a process involving approximately 20 different enzymatic steps. One of the least understood enzymatic steps is formation of the isocyclic ring, which is a characteristic feature of all chlorophyll molecules. In chloroplasts this is an aerobic reaction catalyzed by Mg-protoporphyrin IX monomethyl ester cyclase. Barley mutants were employed to study this enzyme (Rzeznicka et al. 2005). An *in vitro* assay for the aerobic cyclase reaction required both membrane-bound and soluble components from the chloroplasts. Extracts from barley mutants at the *Xantha-l* and *Viridis-k* loci showed no cyclase activity. Fractionation of isolated plastids by Percoll gradient centrifugation showed that both *xantha-l* and *viridis-k* mutants are defective in

components associated with chloroplast membranes. The evidence suggests that the aerobic cyclase requires at least one soluble and two membrane-bound components. The *Xantha-l* gene was located to the short arm of barley chromosome 3H. The gene was further cloned and sequenced and the mutations *xantha-l.*35, *-l.81* and *–l.*82 were characterized at the DNA level. The study connected for the first time biochemical and genetic data as it demonstrated that *Xantha-l* encodes a membrane-bound cyclase subunit.

The stock list and genetic information presented in the Barley Genetics Newsletter 21: 102-108 is valid and up-to-date. Requests for stocks available for distribution are to be either sent to:

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Nordic Gene Bank
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Phone: +46-40-536640
FAX: +46-40-536650
www.nordgen.org/ngb

Reference:

Rzeznicka, K., C. J. Walker, T. Westergren, G. C. Kannangara, D. von Wettstein, S. Merchant, S. P. Gough, and M. Hansson. 2005. *Xantha-l* encodes a membrane protein subunit of the aerobic Mg-protoporphyrin IX monomethyl ester cyclase in the chlorophyll biosynthetic pathway. Proc. Natl. Acad. Sci. USA 102:5886-5891.

Coordinator's report: Ear morphology genes

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No new research on gene localization or descriptions on different morphological genes have been reported since the latest reports in Barley Genetics Newsletter (BGN) or in the AceDB database for Barley Genes and Genetic Stocks.. All descriptions made in the BGN volumes 26, 28, 29, 32 and 35 are still up-to-date and valid. The databases of the Swedish Ear morphology genes are currently updated and will be searchable within International European databases in the future. All different types and characters with its many alleles of the Swedish ear morphology genes are found in the SESTO database of the Nordic Gene Bank. Also, a survey list of the different Swedish ear morphology genes are published in the last volume of Barley Genetics Newsletter, BGN 35:150-154. As my possibilities in searching literature are very limited, I apologize if I am missing any important reports or papers. I would like to call on the barley community to assist me by sending notes of publications and reports to include in next year's reports. Descriptions, images and graphic chromosome map displays of the Ear morphology genes are also available in the AceDB database for Barley Genes and Barley Genetic Stocks. They get currently updated and are searchable under the address: www.untamo.net/bgs

Every research of interest in the field of Ear morphology genes can be reported to the coordinator as well. Seed requests regarding the Swedish mutants can be forwarded to the coordinator udda@ngb.se or udda@nordgen.org or to the Nordic Gene Bank, www.nordgen.org/ngb. all others to the Small Grain Germplasm Research Facility (USDA-ARS), Aberdeen, ID 83210, USA, anhang@uida.edu or to the coordinator at any time.

Coordinator's report: Semidwarf genes

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Dahleen *et al*. (2005) studied 27 mutants from various sources that were placed in the brachytic (*brh*) group of semidwarf mutants. The mutants were backcrossed into 'Bowman' prior to this study to facilitate allelism studies and their phenotypic characterization. The traits studied included plant height; awn, peduncle, and rachis internode length; leaf width and length; lodging; kernels per spike; grain yield; and kernel weight. Based on allelism tests and molecular mapping studies using simple sequence repeat (SSR) markers, the mutants occurred at 18 different loci. Eight of the loci had been identified in previous studies and ten were new loci. Using small F2 populations, SSR markers were mapped within 30 cM of all loci except the *brh15.u* mutant. The brachytic mutants were located as follows: *ert-t* (*brh3.y*), *brh4.j*, and *brh10.l* on chromosome 2H; *brh8.ad* and *brh14.q* on 3H; *brh2* (*ari-l.3*), *brh5.m*, and *brh9.k* on 4H; *brh6.r*, *brh7.w*, *brh11.o*, *brh12.p*, *brh17.ab*, and *brh18.ac* on 5H; and *brh1.z* and *brh16.v* on 7H. The positional information suggested that one or two clusters of brachytic loci may exist on 5H. Three of five loci that were positioned earlier by linkage drag (Franckowiak, 1995) were found in a similar position base on the SSR mapping data.

All of the *brh* mutants as evaluated in Bowman backcross-derived lines were shorter than Bowman with an average height of 64.8 cm vs. 87.9 for Bowman (Dahleen *et al*. 2005). All of the *brh* lines had shorter awns and most had shorter peduncles and smaller kernels. Some of the *brh* lines had shorter rachis internodes and short leaf blades. The majority of the *brh* lines, 16 of 27, had lower grain yields than Bowman. Although none of the *brh* lines was superior to Bowman, the *brh4*, *brh6*, and *brh8* mutants seemed to be the most promising ones for further agronomic evaluation.

Horsley *et al*. (2006) reported that the main plant height QTLs in a 'Foster'/CIho 4196 mapping population were near the *vrs1* locus on 2HL. Dahleen *et al*. (2003) reported a plant height QTL in the same region of 2H from a study of two- by six-rowed cross, ND9712//Foster/Zhedar 2. The association between plant height and the six-rowed phenotype was first reported as a linkage by Miyake and Imai in 1922 and has been reported often since then (Franckowiak 1997). The locus symbol *hcm1* is currently recommended. Horsley *et al*. (2006) provided some evidence that more than one factor for reduced plant height is associated with the *vrs1* locus in the Foster/CIho 4196 cross. They reported also that they did not recover any short plants with a two-rowed spike type from a large F2 population. Thus, it is still not clear whether the *hcm1* locus exists or the six-rowed allele (*vsr1.a*) at *vrs1* locus has a pleiotropic effect on plant height in warm environments.

Honda *et al*. (2003) found that treatment of barley near-isogenic lines with the brassinosteroid (BR) growth regular caused leaf blade rolling in normal barley and most barley semidwarf mutants. However, detached leaf blade segments from dark grown plants with the *uzu1* gene did not unroll after treatment in the leaf unroll test. In a subsequent study, Chono *et al*. (2003) demonstrated that the response of *uzu1* mutants to BR was caused by a base pair substitution in the *Hordeum vulgare* BR-insensitive 1 (*HvBR11*) gene and an amino acid change in a

highly conserved residue in the kinase domain of the BR-receptor protein. The *uzu1* lines have a missense mutation in the *HvBR11* gene.

Gottwald *et al*. (2004) reported that a gibberellic-acid insensitive dwarf mutant, first described by Favret *et al*. (1976), is closely linked to RFLP marker MWG2287 on 2HS near the centromere. The proposed locus symbol for the GA insensitive mutant is *sdw3*, which replaced the symbols *gai* and *GA-ins* used in earlier publications. The suggested allele symbol is *sdw3.az* for the Hv287 line derived from the M.C. 90 mutant induced in M.C. 20. This region of 2HS is orthologous with a highly conserved region on rice chromosome 7L. ESTs in this region were used to identify three putative GA-related ORFs in rice that might correspond to the *sdw3* locus. (Gottwald *et al*. 2004).

References:

Chono, M., I. Honda, H. Zeniya, K. Yoneyama, D. Saisho, K. Takeda, S. Takatsuto, T. Hoshino, and Y. Watanabe. 2003. A semidwarf phenotype of barley *uzu* results from a nucleotide substitution in the gene encoding a putative bassinosteroid receptor. Plant Physiol. 133:1209-1219.

Dahleen, L.S., H.A. Agrama, R.D. Horsley, B.J. Steffenson, P.B. Schwarz, A. Mesfin, and J.D. Franckowiak. 2003. Identification of QTLs associated with Fusarium head blight resistance in Zhedar 2. Theor. Appl. Genet. 108:95-104.

Dahleen, L.S., L.J. Vander Wal, and J.D. Franckowiak. 2005. Characterization and molecular mapping of genes determining semidwarfism in barley. J. Hered. 96:654-662.

Favret, E.A., G.C. Favret, and E.M. Malvarez. 1976. Genetic regulatory mechanisms for seedling growth in barley. p. 37-41. p. 181-189. *In* H. Gaul (ed.). Barley Genetics III. Proc. Third Int. Barley Genet. Symp., Garching, 1975. Verlag Karl Thiemig, München.

Franckowiak, J.D. 1995. Notes on linkage drag in Bowman backcross derived lines of spring barley. BGN 24:63-70.

Franckowiak, J.D. 1997. BGS 77, short culm, *hcm*, revised. BGN 26:115.

Gottwald, S., N. Stein, A. Börner, T. Sasaki, and A. Graner. 2004. The gibberellic-acid insensitive dwarfing gene *sdw3* of barley is located on chromosome 2HS in a region that shows high colinearity with rice chromosome 7L. Mol. Gen. Genomics 271:426-436.

Honda, I., H. Zeniya, K. Yoneyama, M. Chono, S. Kaneko, and Y. Watanaba. 2003. *Uzu* mutation in barley (*Hordeum vulgare* L.) reduces leaf unrolling response to brassinole. Biosci. Biotechnol. Biochem. 67:1194-1197.

Horsley, R.D., D. Schmierer, C. Maier, D. Kudrna, C.A. Urrea, B.J. Steffenson, P.B. Schwarz, J.D. Franckowiak, M.J. Green, B. Zhang, and A. Kleinhofs. 2006. Identification of QTLs associated with Fusarium head blight resistance in barley accession CIho 4196. Crop Sci. 46:145-156.

Coordinator's report: Early maturity genes

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No new research on gene localization has been reported on the Early maturity or Praematurum genes since the latest reports in Barley Genetic Newsletter (BGN) or in the AceDB database for Barley Genes and Barley Genetic Stocks. All information and descriptions made in the Barley Genetics Newsletter are valid and up-to-date. As my possibilities in searching literature are very limited, I apologize if I am missing any important papers and reports. I would like to call on the barley community to assist me by sending notes of publications and reports to include in next year's report. Descriptions, images and graphic chromosome map displays of the Early maturity or Praematurum genes are available in the AceDB database for Barley Genes and Barley Genetic Stocks. They get currently updated and are searchable under the address: www.untamo.net/bgs

Every research of interest in the field of Early maturity genes can be reported to the coordinator as well. Seed requests regarding the Swedish mutants can be forwarded to the coordinator or directly to the Nordic Gene Bank, www.nordgen.org/ngb, all others to the Small Grain Germplasm Research Facility (USDA-ARS), Aberdeen, ID 83210, USA, anhang@uidaho.edu or to the coordinator at any time.

Coordinator's report : Wheat-barley genetic stocks

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The production of five different monosomic addition lines of *Hordeum marinum* chromosomes to Chinese Spring wheat has been reported earlier. It has now been possible to isolate five disomic addition lines (1Hm, 2Hm, 4Hm, 5Hm and 7Hm) from them and work is in progress to isolate the two remaining (3Hm and 6Hm) addition lines. Apart from the production of *H. marinum* x CS wheat amphiploid, it has also been possible to produce amphiploid with commercial wheats, both common and durum (Islam and Colmer, unpublished).