



Characterisation of mapping parents and identification of genes involved in the yield and grain weight of barley (*Hordeum vulgare* L.) grown under Mediterranean environments

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Introduction

The yield of barley is reduced by drought and heat stress during crop development, which commonly occurs under Mediterranean environments (Van Oosterom and Acevedo 1992). Many potential traits have been suggested to improve yield stability under stressful environments, however relatively few have lead to yield stable genotypes. Early maturity and the completion of grain-filling before heat and drought stress is yield limiting but has been the main trait selected to breed genotypes suitable for Mediterranean environments (Ceccarelli *et al.* 1991; Shakhathreh *et al.* 2001). Still, improvement of yield stability has been slow because stressful environments lead to large genotype x environment (GxE) effects, making heritability for yield small, and selection of superior genotypes difficult. To improve the yield of barley under Mediterranean environments, an understanding of the genes determining traits involved in yield under stress is necessary.

Materials and Methods

Sowing Date Studies

Various barley genotypes were grown at multiple sowing dates to induce progressively more stressful growing conditions. In 1999 and 2000 field trials were conducted at the Charlick Experimental Station (Strathalybn). These were split-plot designs with four replicates as blocks, three sowing dates as whole plots, and varieties as sub plots in the 1999 experiment, and the same varieties in 2000 but with two replications and six sowing dates. The sowing dates for 1999 were 28/06/99, 16/07/99, 11/08/99, and for 2000 they were 20/6/00, 3/7/00, 15/7/00, 1/8/00, 21/8/00, 6/9/00. The traits measured were growth habit, early vigour, Zadoks scores (Decimal Growth Stage; DGS), lodging, plant height, spikelet mortality, anthesis date, harvest

index, physiological maturity, grain weight, grain number, screenings (<2.2mm), and plot yield. The data was spatially analysed using REML analysis in Genstat 5.

Mapping Population Studies

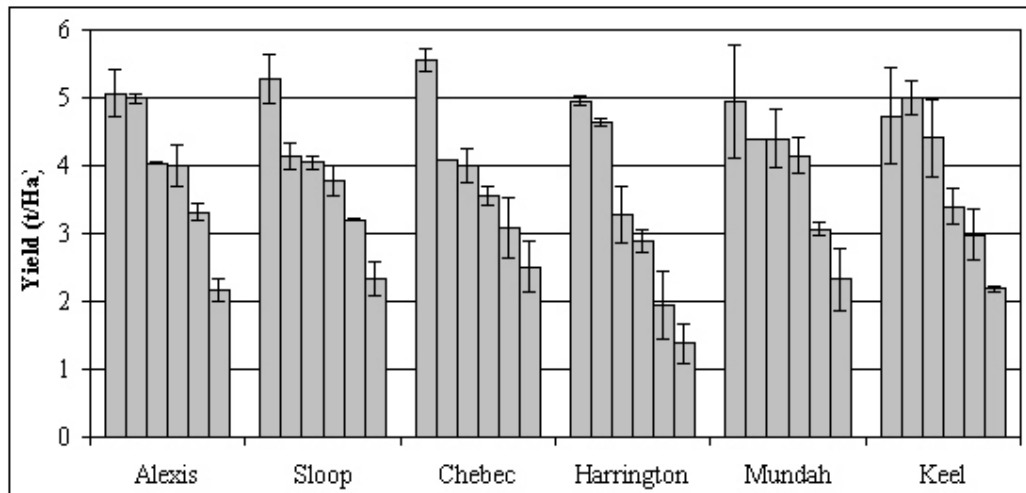
The mapping populations Alexis x Sloop (AxS) and Chebec x Harrington (CxH) were used in a QTL mapping approach to identify genes involved in the yield of barley under stress. These populations were grown at the low rainfall site of Pt. Wakefield. The 1999 trials were unreplicated with the parents in a regular grid, and with two replications in 2000. The new mapping population Mundah x Keel (MxK) was also grown at Pt. Wakefield in 2000. The traits measured were establishment, growth habit, early vigour, DGS, height, head loss, lodging, grain number, tiller number, biomass, harvest index, grain weight, screenings, and plot yield. AxS was grown in the summer nursery in 2001 and was additionally evaluated for anthesis date, chlorophyll (SPAD), primary tiller leaf number, and flag leaf area, leaf dry weight, and specific leaf area. The AxS population was grown at two sowing dates in 2000 at Charlick, each with two replications, and was evaluated for development, height, grain weight, and yield.

Results and Discussion

Sowing Date Studies

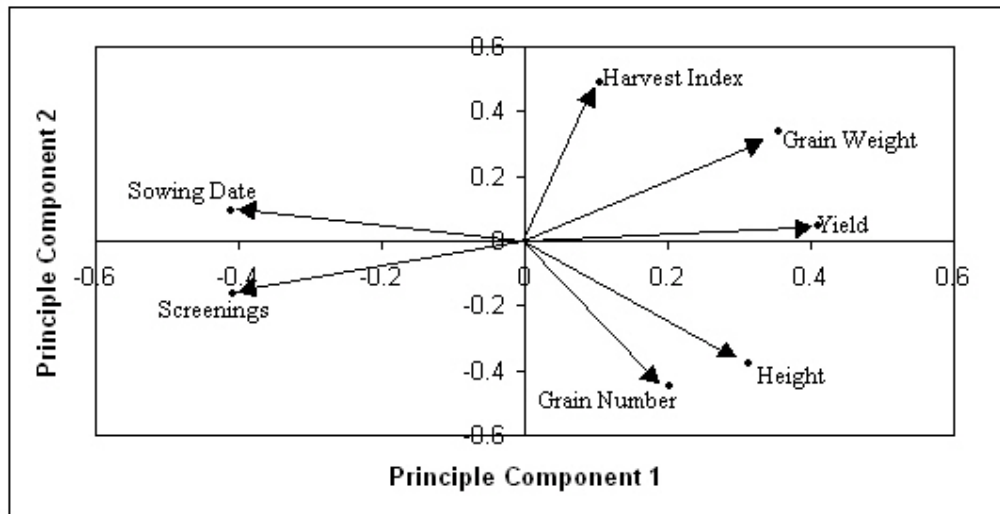
The yield of all cultivars progressively declined with sowing date-induced stress in 1999 and 2000. This was especially noticeable in the unadapted mapping parents Alexis and Harrington (Figure 1), and similarly Gairdner and Franklin (not shown). Sloop, Chebec, Mundah and Keel maintained more stable yields (less change over sowing dates).

Figure 1: The yield response of six mapping parents to six different sowing date environments in a field trial grown at Charlick in 2000 (sowing dates 1 to 6 from left to right for each genotype)



The correlation structure of a set of variables measured in the sowing date trials was analysed by principle components analysis (PCA). The traits explaining variability in genotype mean yield across sowing dates is shown by the PCA biplot (Figure 2) where uncorrelated variables are at 90° . The components with the highest correlations with yield were sowing date (-0.9), individual grain weight (0.68), and screenings (-0.85). This indicates grain yield at different sowing dates is strongly related to grain weight. The yield trends were reflected in individual grain weight (Figure 3) in terms of decrease with sowing date, and maintenance of grain weight over sowing dates for stable genotypes. Harrington appears to have particularly unstable grain weight compared to Chebec across all sowing dates, and there is significant variation between Alexis and Sloop under the later sowing dates. This indicates that mapping these traits would detect QTL for yield and grain weight under favourable and stressful conditions.

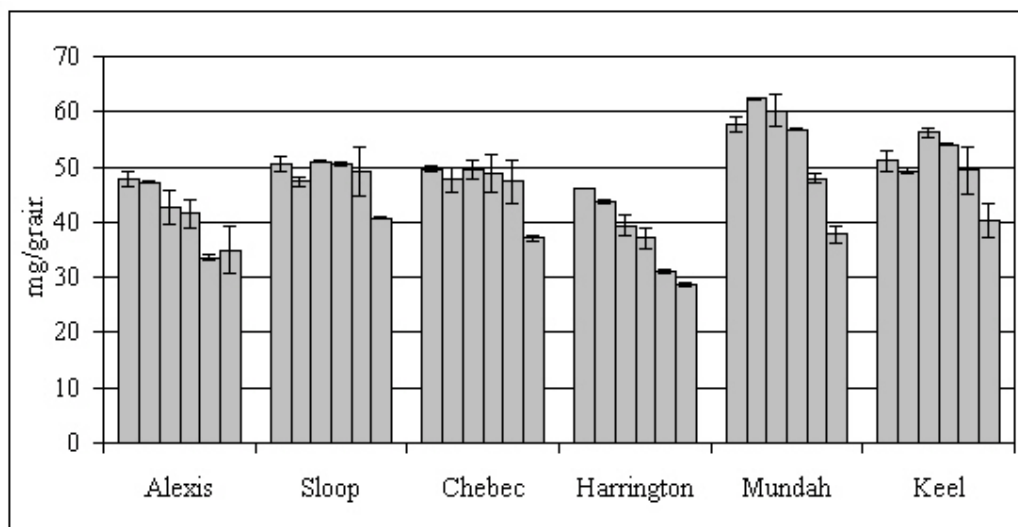
Figure 2: PCA biplot summarizing the relationships among variables of the sowing date trial conducted at Charlick in 2000.



Mapping Population Studies

Loci affecting yield and yield related traits on chromosomes 2H, 3H, and 7H in the AxS and CxH mapping populations were determined. Locations of QTL for traits measured in MxK will be determined on completion of its genetic map. Two loci are found on chromosome 2H, and include the photoperiod response gene *Ppd-H1*, linked to *Xabg2* and the STS marker *Xpsb992* (Decousset *et al* 2000) mapped in AxS, and another locus identified by the marker *Xabg14*. Under the 1999 drought conditions, *Ppd-H1* was coincident with QTL for early vigour and height in AxS, and screenings in CxH. The locus *Xabg14* was coincident with QTL for DGS, head loss, yield, and thousand grain weight in AxS (Table 1) and CxH. This locus remained to exert its effects on thousand grain weight, plump grain, and screenings under the favourable conditions of 2000 in AxS. When the 1999 yield data was adjusted using DGS as a covariate, the *Xabg14* locus had a non-significant LOD score. The locus *Xabg14* is important in determining yield and grain weight under drought and favourable conditions through its effects on barley development.

Figure 3: The response of the individual grain weight of six mapping parents to six different sowing date environments in a field trial grown at Charlick in 2000



Boyd (unpublished data) mapped juvenile stage and awn appearance under extended and normal photoperiods in CxH, and found the *Ppd-H1* (*Xabg2*) locus was responsive to increasing photoperiod, whilst the other 2H locus (*Xabg14*) was associated with QTL for these traits under normal photoperiod. To further quantify the effects of these loci in AxS, the population was grown in a summer nursery under irrigation, extended photoperiod, and heat stress. Under these conditions the *Ppd-H1* locus was strongly associated with traits related to development and photosynthesis, whilst *Xabg14* was less strongly associated but still significant (Table 1). Under sowing date induced stress these loci had no detectable effects on yield and thousand grain weight in AxS. The 2H loci have been associated with QTL for developmental and agronomic related traits in other studies (Backes *et al.* 1995; Bezant *et al.* 1996; Hayes *et al.* 1993; Laurie *et al.* 1995; Karsai *et al.* 1997; Karsai *et al.* 1999).

The major stature gene *sdw1* (syn. *sdw1*; *denso*) has been mapped on chromosome 3H (Laurie *et al.* 1993) and segregates in the AxS population. This locus is detected by *Xabg4* and displayed effects on the yield related traits measured in 2000 under favourable conditions and in the sowing date experiment, including thousand grain weight, individual grain weight, screenings, harvest index, and yield. It had strong effects on early vigour, growth habit, DGS, and plant height in all experiments except the summer nursery experiment. The *sdw1* gene has been shown to have major effects on many traits in other mapping populations (Barua *et al.* 1993; Laurie *et al.* 1995; Benzant *et al.* 1996; Yin *et al.* 1999).

Table 1. A graphical summary of the Alexis x Sloop mapping data showing significant QTL (shaded; LOD>3) associated with various traits evaluated under different environments

Trait	3H	2H	2H	7H	7H
	Sdw1	Ppd-H1	Xabg14	Xbmac755	XsssI
	Xabg4	Xabg2			

Pt Wakefield 1999 Drought and Heat Stress	Early Vigour	■	■			■	
	Growth Habit	■					
	Height	■	■				
	DGS	■		■			
	Head Loss			■			
	Yield			■			
	1000-Grain Weight			■	■		
Pt Wakefield 2000 Favourable Conditions	IGW	■					
	Plump	■		■			
	Screenings	■		■			
	Tiller Weight	■					
	Habit	■					
	DGS	■	■	■			
	Early Vigour	■					
	1000-Grain Weight	■		■			
	Harvest Index	■					
	Height	■	■				
	Lodging					■	
	Charlick 2000 Sowing Date Trial	1000-Grain Weight SD1	■			■	
		Yield SD1	■				
Height SD2		■					
1000-Grain Weight SD2		■			■		
DGS SD2			■	■			
Yield SD2					■		
2001 Summer Nursery Heat stress and long photoperiod	Anthesis date		■	■			
	Biomass		■	■			
	Leaf Area		■	■			
	Leaf Number		■	■			
	Leaf Weight		■	■			
	No. Tillers		■	■			
	Chlorophyll (SPAD)		■	■			
	Specific Leaf Area		■				
	Tiller Weight		■				

In the sowing date experiment a 7H locus (*Xbac755*) was mapped for yield and thousand grain weight. Its is coincident with a leaf rust QTL, and the presence of leaf rust in this experiment may explain the association with this locus by the influence of leaf rust on the traits measured. A second 7H locus (*Xsss1*) was detected for early vigour and lodging in AxS, and head loss in CxH. This also coincides with a minor QTL for awn appearance under normal photoperiod, and probably indicates another locus effecting development.

Conclusion

The detection of loci influencing yield, and their association with QTL for yield determinants, will allow a genetic dissection of this complex trait into more understandable components. This will improve breeding for yield under stress through developing genotypes carrying important simply inherited traits related to yield. The results presented here show that plant development is an important adaptation mechanism to stressful environments. Identification of genes involved in yield under stress independent of plant development would be valuable to breeding as later maturing genotypes would maintain yield potential under favourable conditions, and a yield advantage under stressful conditions. The strong effects of the photoperiod response genes in both populations, and the *sdw1* gene in AxS obscured the detection of novel genes influencing yield and grain weight. A more thorough assessment of the loci influencing development and their contribution to GxE interactions is needed before novel genes/QTL involved in yield and grain weight under challenging environmental conditions can be found. Accurate phenotyping of populations to locate QTL, considering the large GxE interactions under stressful environments is difficult, but once identified will truly be amendable to a marker assisted selection approach to improve yield and grain weight stability.

Acknowledgments

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References

1. Backes, G., Graner, A., Foroughi-Wehr, B., Fischbeck, G., Wenzel, G., and Jahoor, A. (1995) Theor. Appl. Genet., 90:294-302.

2. Barua, U.M., Chalmers, K.J., Thomas, W.T.B., Hackett, C.A., Lea, V., Jack, P., Forster, B.P., Waugh, R., and Powell, W. (1993) *Genome.*, 36:1080-1087.
 3. Bezant, J., Laurie, D., Pratchett, N., Chojecki, J., and Kearsley, M. (1996) *Heredity.*, 77:64-73.
 4. Ceccarelli, S., Acevedo, E., and Grando, S. (1991) *Euphytica.*, 56:169-185.
 5. Decousset, L., Griffiths, S., Dunford, R.P., Pratchett, N., and Laurie, D.A. (2000) *Theor. Appl. Genet.*, 101:1202-1206.
 6. Hayes, P.M., Liu, B.H., Knapp, S.J., Chen, F., Jones, B., and Blake, T. (1993) *Theor. Appl. Genet.*, 87: 392-401.
 7. Karsai, I., Meszaros, K., Szucs, P., Hayes, P.M., Lang, L., and Bedo, Z. (1999) *Plant Breed.*, 118:399-403.
 8. Karsai, I., Meszaros, K., Hayes, P.M., Bedo, Z. (1997) *Theor. Appl. Genet.* 94: 612-618.
 9. Laurie, D.A., Pratchett, N., Romero, C., Simpson, E., and Snape, J.W. (1993) *Plant Breed.*, 111:177-264.
 10. Laurie, D.A., Pratchett, N., Bezant, J.H., Snape, J.W. (1995) *Genome* 38:575-585.
 11. Shakhathreh, Y., Kafawin, O., Ceccarelli, S., and Saoub, H. (2001) *J. Agronomy & Crop Science.*, 186:119-127.
 12. Van Oosterom, E.J., and Acevedo, E. (1992) *Euphytica.*, 62:29-38.
 13. Yin, X., Stam, P., Dourleijn., C.J., and Kropff., M.J. (1999) *Theor. Appl. Genet.*, 99(1-2):244-253.
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