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Detection of quantitative trait loci for agronomic, yield, grain and disease characters in spring barley (*Hordeum vulgare* L.)

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Abstract Quantitative trait loci (QTLs) have been revealed for characters in a segregating population from a spring barley cross between genotypes adapted to North-West Europe. Transgressive segregation was found for all the characters, which was confirmed by the regular detection of positive and negative QTLs from both parents. A QTL for all the agronomic, yield and grain characters measured except thousand grain weight was found in the region of the *denso* dwarfing gene locus. There were considerable differences between the location of QTLs found in the present study and those found in previous studies of North American germ plasm, revealing the diversity between the two gene pools. Thirty-one QTLs were detected in more than one environment for the 13 characters studied, although many more were detected in just one environment. Whilst biometrical analyses suggested the presence of epistasis in the genetic control of some characters, there was little evidence of interactions between the QTLs apart from those associated with yield. QTLs of large effect sometimes masked the presence of QTLs of smaller effect.

Key words Barley · QTLs · Linkage · Yield · Markers

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Introduction

The improvement of crop plants is usually achieved through crossing parents that complement each other for desirable characters, followed by the selection of appropriate recombinants. In barley most of the economically important characters are quantitative, requiring replicated field trials for accurate measurement. As this is time-consuming and expensive, breeders are looking for more rapid alternatives to the conventional selection schemes. The use of markers to identify and select individual loci controlling quantitative traits (QTLs) offers one such possibility.

The efficient deployment of markers in breeding and research requires the identification of robust associations between a marker and a trait. Morphological markers have been used to identify associations with some traits in barley (Suneson and Stevens 1957); however, such markers are often deleterious and their application in plant breeding has been limited. The development of biochemical markers led to the identification of associations with some major genes (Forster et al. 1991), but these markers have not been widely deployed due to their infrequency and/or monomorphism in adapted germ plasm. DNA-based markers, such as restriction fragment length polymorphisms (RFLPs) and randomly amplified polymorphic DNA (RAPDs) have been developed over the last 10 years and appear to suffer none of the disadvantages of morphological and biochemical markers. RFLP, in particular, has led to the creation of molecular genetic linkage maps in a number of species, including four in barley (Heun et al. 1991; Graner et al. 1991, 1994; Kleinhofs et al. 1993, 1994; Kasha et al. 1994). In addition, a genetic map of barley has been published that is largely based on RAPDs (Giese et al. 1994).

Associations between markers and traits of interest can be revealed from studies based upon measurements of the trait in mapped populations. Such studies have led to the mapping of previously unassigned qualitative loci

in barley such as the *denso* dwarfing gene (Barua et al. 1993b; Laurie et al. 1993), the *Ml(La)* mildew resistance (Hilbers et al. 1992; Giese et al. 1993) and the *amo1* high amylose gene (Schondelmaier et al. 1992). The barley maps have also been used to locate QTLs for single traits such as quantitative powdery mildew resistance (Heun 1992), milling energy (Chalmers et al. 1993), cold hardiness (Hayes et al. 1993a) and heading date and height (Barua et al. 1993b). Quantitative trait loci for a number of characters have been studied in two crosses between North American spring barley cultivars (Hayes et al. 1993b; Han and Ullrich 1994; Tinker et al. 1994). Backes et al. (1995) report the location of QTLs for some agronomic traits and yield in a cross between two winter barley cultivars. In the present article we report the use of a genetic linkage map of the barley genome to reveal QTLs for agronomic, yield, grain and disease characters scored in 4 years of field trials. This approach should maximise the environmental component of the characters and therefore enable us to locate regions of the genome with which they are consistently associated.

Materials and methods

Traits

Fifty-nine doubled haploid (DH) lines derived from a spring barley cross between 'Blenheim' and the Scottish Crop Research Institute (SCRI) breeding line E224/3 (Finnie et al. 1989) were grown together with their parents in a replicated trial at SCRI each year from 1989 to 1992 and at Plant Breeding International Cambridge (PBIC) in 1992 and 1993. The plots were sprayed with fungicides at an average growth stage, (GS)31 (Tottman 1987) and GS39, to provide protection from foliar pathogens. During the growing season, plots at SCRI were scored for heading date (HD), the number of days after May 31st on which 50% of the plot had reached GS53, and for height (Ht), cm from the ground to the collar at GS85. When ripe, the plots were harvested with a small plot combine and the seed dried to 13% moisture before evaluating plot yield (PY) (t/ha). The seed from each plot was then cleaned and specific weight (SPW) measured using a chondrometer. A sample of cleaned seed was passed over 2.8-, 2.5- and 2.2-mm sieves, and each retained fraction was, for the 2.5- and 2.2 mm sieves, amalgamated with larger fractions and expressed as the angular transformation of a percentage of the whole sample (>28, >25 and >22, respectively). The amalgamated fractions greater than 2.5 mm were used for all subsequent measurements. Thousand grain weight (TGW) was calculated from the weight of 100 grains. At PBIC, PY was recorded in 1992 and 1993 and TGW in 1992 only.

In addition, the DH lines and parents were grown in unreplicated disease nurseries for mildew, *Rhynchosporium* and brown and yellow rust at the Welsh Plant Breeding Station in 1990 and their reaction to the diseases scored twice (1M1, 2M1, 1Rh, 2Rh, 1BR, 2BR, 1YR and 2YR). The first score was carried out at approximately GS53, and the second, 2 weeks later for mildew. For the other diseases, the first score was carried out at approximately GS59, and the second, 10 days later. The lines were also grown in a replicated *Rhynchosporium* nursery at SCRI in 1991 and 1992 and scored twice (1Rh and 2Rh) and in a replicated mildew nursery at SCRI in 1992 and scored once (1M1). Disease was spread from natural infection in the mildew and *Rhynchosporium* nurseries and from innoculation with Triumph virulent races in the rust nurseries. Whilst the Scald scores were used to categorise the DH lines as resistant or susceptible and place the resistance locus on our linkage map (Barua et al. 1993a), it was obvious from the scores that considerable variation existed in the resistant and susceptible groups, and the percentage scores for 1Rh and 2Rh were therefore treated as quantitative traits.

Markers

Seventy-five RAPD (coded SC, OP or R) and 54 RFLP (coded ABG, BCD, BG, CDO, G, MWG, PBI, WG or PSR) polymorphic markers were scored on the DH lines by the protocols of Barua et al. (1993b). Five biochemical markers (EST1, EST2, AMY1, WSP2 and WSP5), a sequence tagged site (*Ale*), one morphological marker (*denso*) and *Rhynchosporium* resistance (*Rh*) were also scored on the lines. The biochemical markers were scored by the protocols of Thompson et al. (1990). The sequence-tagged site was scored by amplification of a fragment of the aleurain sequence (Li et al. 1991). The *denso* gene confers a semi-prostate growth habit that was scored in trials at GS24 from 1989 to 1992. Scald resistance was scored as percentage leaf area infected in replicated disease nurseries grown in 1991 and 1992. A major *Rynchosporium* resistance gene segregated in the DH population and the percentage scores were used to classify lines as resistant or susceptible by comparison with the parental scores (Barua et al. 1993a).

The ordering of the markers was investigated with MAPMAKER (Lander et al. 1987) and JOINMAP (Stam 1993) using the Haldane mapping function.

Analysis

The location of QTLs controlling the traits was examined by stepping down the linkage groups at intervals of 2% recombination and fitting an expected additive genetic effect (Haley and Knott 1992). The location that gave the most significant regression was taken as the most likely position of a QTL, rather then the likelihood ratio test (Haley and Knott 1992), which we found to be more sensitive to missing data values. In addition to the linkage groups, the unlinked markers were also considered in the analysis. A stepwise procedure was adopted in screening for further QTLs for a character. The first QTL was fixed and the most likely position of a second QTL was determined as above with its significance determined as described (Haley and Knott 1992). This process was repeated for up to 6 QTLs or until a QTL was not significant at the 5% level. Once the positions of QTLs were established, the expected additive-additive epistatic effect for each pair of QTLs was calculated (Haley and Knott 1992) and the STEP directive in GENSTAT used to identify significant interactions.

Results

Markers

Twenty-four RAPD markers were excluded from the mapping analysis as they either had many missing scores or their inclusion produced inconsistencies in a linkage group. A LOD of 3.0 for linkage was found to produce reasonably tight linkage groups that, with four exceptions, contained specific markers that mapped to the same chromosome. There were some differences between JOINMAP and MAPMAKER in the ordering of loci in a few groups, but JOINMAP produced substantially shorter maps for many of the linkage groups and the orderings were judged to be robust, as they did not vary with alterations in the maplod score (Stam 1993). Maps produced by JOINMAP were therefore used in all subsequent analyses. One hundred and eight of the markers were assigned to linkage groups, with 6 RFLPs and 6 RAPDs not showing significant linkage with any other marker. Groups were assigned to chromosomes when they contained previously located chromosome-specific RFLPs, and the most likely ordering of

groups was determined by the comparison of common markers with the published maps of Heun et al. (1991), Graner et al. (1994) and Kleinhofs et al. (1994). Orderings within groups were generally consistent with those shown on previous maps with the exception of BCD98 and BCD249 relevant to the Hordein loci (Heun et al. 1991; Sogaard and von Wettstein-Knowles 1987). Four linkage groups were found covering 150 cM of chromosome 1 (7H), 2 groups covered 110 cM chromosome 2 (2H), 3 covered 180 cM of chromosome 3 (3H), 1 covered 15 cM of the short arm of chromosome 4 (4H), 2 covered 50 cM of chromosome 5 (1H), 3 covered 75 cM of chromosome 6 (6H), 2 covered 95 cM of chromosome 7 (5H) and there was 1 small linkage group that could not be assigned to any chromosome that covered 15 cM (Fig. 1).

Biometrical analysis

There was transgressive segregation in both directions for all characters in all environments as the highest and lowest DH lines were always higher and lower than the higher and lower scoring parent, respectively (Tables 1 and 2). This suggests that each parent possessed positive and negative alleles for QTLs controlling the characers studied. Substantial evidence for this is provided by the regression analysis, which reveals positive and negative QTLs from 'Blenheim' in 32 of the 48 character and environment combinations (Tables 1 and 2). In the remainer, either only 1 QTL was detected or all of the positive alleles came from one parent. In the latter category there were only 4 instances where 3 or more QTLs were detected. Apart from TGW, where increasing QTLs from E224/3 were detected in just one of the five environments, there was no pattern in the instances where all of the increasing QTLs were derived from one parent. There were some indications of epistatsis (Tables 1 and 2) as the DH mean was occasionally found to differ significantly from the mid-parental value (Snape and Simpson 1981a). This occurred twice for PY and was also significant at the 10% level at PBIC in 1993. There was also evidence of epistatis in more than one environment for 2Rh. There were a number of other instances where the DH mean fell outside the parental range, but the differences between the parents were generally small in such cases and epistatis was not significant.

QTL mapping

The ratio of the difference between the highest and lowest scoring inbred line to the additive genetic vari-

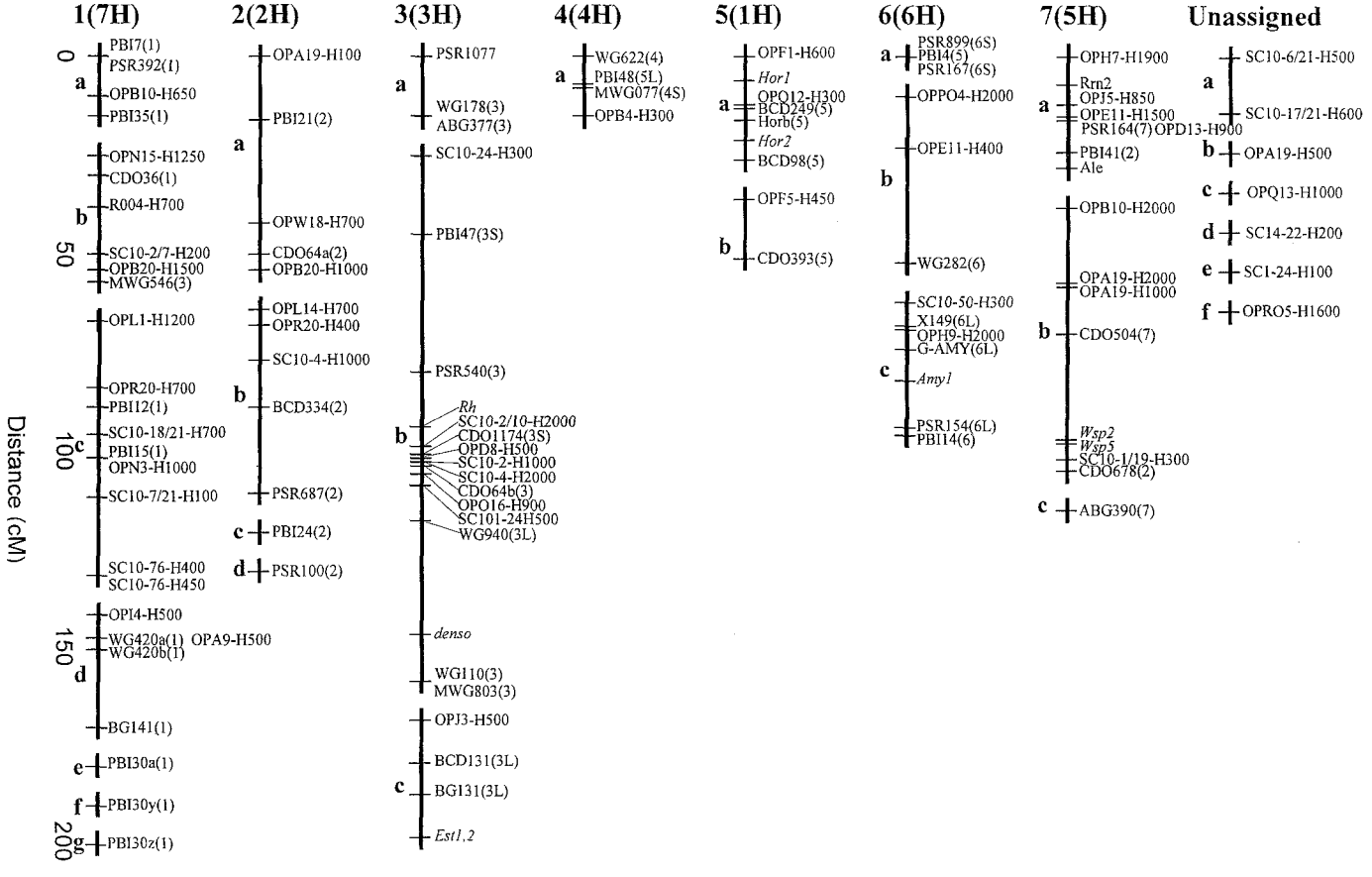

Fig. 1 Chromosome maps for 'Blenheim' × E224/3 doubled haploid population. Marker location is indicated by a *horizontal line* and groups linked at LOD 3.0 are joined by a *vertical bar*. Individual segments of a chromosome are arranged and lettered in their most likely order by comparison with previously published maps and unlinked chromosome specific markers are arranged at chromosome ends. Protein, isozyme, morpohological and disease resistance loci are *italicised*

Table 1 Performance of parents and DH lines, percentage variation accounted for and numbers of effective factors (k) and of increasing (+) and decreasing (−) QTLs from 'Blenheim' detected for agronomic, yield and grain characters by regression

Character	Year[a]	Blenheim	E 224/3	DH Lines			k	%Variation	Blenheim alleles	
				Maximum	Mean[b]	Minimum			+	−[c]
HD	89	20.50	21.50	27.00	21.85	19.00	4.46	45.5	1	1
HD	90	25.00	24.00	29.00	25.46	21.00	–	72.3	3	2
HD	91	24.75	25.00	29.50	25.53	22.00	3.02	85.9	2	4
HD	92	15.50	17.00	24.00	**17.26**	13.50	6.93	86.1	2	4
Ht	89	55.50	66.25	77.50	60.35	46.00	3.57	86.2	1	2
Ht	90	72.25	83.75	109.50	79.29	55.00	4.38	92.6	2	2
Ht	91	66.75	76.00	93.50	74.21	57.00	1.89	93.4	1	2
Ht	92	59.75	69.00	84.50	67.53	50.00	3.19	87.2	2	2
PY	89	5.589	4.948	6.203	**4.904**	3.434	5.81	68.4	2	0
PY	90	6.607	7.395	8.309	6.875	4.644	11.96	45.1	1	0
PY	91	7.434	6.623	7.522	**6.471**	5.396	7.24	72.4	2	2
PY	92	7.364	7.635	8.977	7.347	5.211	8.96	64.8	1	1
PY	92C	6.066	5.762	6.355	5.526	3.935	6.88	50.1	**3**	**0**
PY	93C	6.963	6.473	6.986	6.046	4.356	3.49	80.2	**2**	**1**
TGW	89	51.34	46.87	53.24	48.84	44.67	8.27	21.3	1	0
TGW	90	49.83	46.52	52.86	48.10	41.71	6.22	29.1	1	0
TGW	91	51.34	46.85	54.07	49.21	44.09	5.97	47.2	2	0
TGW	92	53.79	49.76	56.26	51.94	45.02	4.75	73.6	2	3
TGW	92C	48.80	43.50	50.00	45.29	37.40	5.83	47.9	2	0
SPW	89	72.34	71.06	74.59	71.15	67.11	10.70	34.7	0	3
SPW	90	70.08	70.69	71.93	69.58	66.14	6.80	42.1	0	2
SPW	91	70.68	70.32	74.11	70.54	67.00	6.77	78.6	**3**	**3**
SPW	92	72.77	72.37	74.74	72.34	68.86	5.43	54.7	1	2
>28	89	48.21	46.49	58.72	45.62	29.66	6.17	77.6	1	5
>28	90	65.36	64.07	72.75	63.66	55.19	4.81	47.9	3	0
>28	91	51.00	50.10	61.71	50.44	34.11	5.09	49.1	1	1
>28	92	59.26	61.84	72.31	59.40	42.70	4.84	52.8	1	2
>25	89	65.38	61.37	69.27	62.67	55.64	7.05	72.0	2	4
>25	90	78.42	77.37	82.77	76.56	68.64	12.39	61.2	3	1
>25	91	70.17	71.30	76.44	70.55	61.92	4.98	53.5	1	1
>25	92	78.01	79.54	83.15	78.36	73.08	5.06	42.8	1	1
>22	89	75.76	71.61	80.16	74.28	69.51	8.64	13.5	0	1
>22	90	83.21	83.35	85.00	82.27	77.23	–	64.9	2	3
>22	91	80.21	80.88	83.88	80.05	74.21	4.97	77.2	3	3
>22	92	85.54	85.64	86.70	85.17	82.67	–	78.6	1	4

[a] C = Measurement at PBIC site; all other measurements at SCRI site

[b] Underlined and bold means significantly differ from mid-parent at 10% and 5% level, respectively

[c] Numbers in bold indicate significant interactions between QTLs.

ance provides an estimate of the number of effective factors (k) segregating in a cross (Mather and Jinks 1983). Estimation of the additive genetic variance was not possible for the characters scored in the unreplicated disease nurseries, and the phenotypic variance was used instead, which would lead to under-estimates of k. In three cases (HD in 1990 and >22 in 1990 and 1992), no significant genetic variation was detected between the DH lines so no estimate of k could be made. In the other 45 character and environment combinations, the greatest value of k was found for >25 in 1990 and the lowest for Ht in 1991. However, considerable variation over environments in the estimates of k and the number of QTLs detected for each character is apparent from Tables 1 and 2. In most cases, estimates of k exceeded the numbers of QTLs detected, but there were 6 cases where the regression analysis exceeded k by at least 1 QTL.

Overall, the QTLs detected accounted for up to 93% of the phenotypic variance in a character (Ht in 1991) and down to 14% (>22 in 1990), with over 50% of the variation being accounted for in most instances (Tables 1 and 2).

Agronomic (HD and Ht), yield (PY) and grain (TGW, SPW, >28, >25 and >22) characters

Each detected QTL is represented by a bar in Fig. 2a–h. The bar is placed at the most likely location of the QTL, and its size represents the additive effect of the 'Blenheim' QTL allele compared to the estimated mean of all inbred lines from the cross. For linked markers, the interval over which the regression coefficient remained significant was taken as an estimate of the confidence

limits of a QTL. If QTLs for a given trait were all positive or all negative and coincident, or if their confidence intervals overlapped over two or more environments, it was concluded that a QTL for the trait was probably located in that region of the genome. Interacting QTLs are denoted by an asterisk but the regression analysis only detected epistasis for PY at Cambridge in 1992 and 1993 and SPW in 1991.

For HD 3 QTLs were detected in more than one environment with two 'Blenheim' alleles on chromosomes 6 and 7, respectively, decreasing HD and one at the *denso* locus on chromosome 3 increasing HD. None of the QTLs were detected in all four environments in which the character was measured, but all three were found in three (Fig. 2a).

Three QTLs were regularly detected for Ht, the 'Blenheim' allele at the *denso* locus consistently decreasing height by an average of 10 cm. However, 2 increasing QTLs from 'Blenheim' were also detected; one was located on a segment of chromosome 1 in all four environments and increased height by an average of 3 cm; the other was located on the long arm of chromosome 7 in two environments and also increased height by an average of 3 cm (Fig. 2b).

Two increasing QTLs from 'Blenheim' for PY were detected on chromosomes 3 and 7, respectively. The one on chromosome 3 was located very close to the *denso* locus in all six environments in which the character was measured and increased PY by an average of 0.5 t/ha. The other was located close to the *Rrn2* locus in three environments and increased PY by an average 0.2 t/ha (Fig. 2c). The QTL in the region of *denso* interacted significantly with that at *Rrn2* in both environments at Cambridge. In both these environments, the DH mean was less than that of the lower yielding parent, although the differences were not significant (Table 1).

Three QTLs from 'Blenheim' that increased TGW were detected in more than one environment. One on chromosome 1, that increased TGW by an average of 1.2 g, was found in each environment except 1990. The other two, which both increased TGW by an average of 1.1 g, were located on chromosomes 3 and 6, respectively, and were detected in two environments (Fig. 2d).

In contrast, all of the QTL alleles from 'Blenheim' that were detected in more than one environment decreased SPW. Two were located on chromosome 3: the one with the greatest effect was detected in all four environments and was located close to the *denso* locus; the other was detected only in two environments and was probably located on the short arm of the chromosome. These 2 QTLs showed a significant interaction from the regression analysis in 1991, although there was little difference between the DH mean and mid-parental value. The third QTL was detected in three environments and was located in the region of the *Amy1* locus on chromosome 6 (Fig. 2e).

Increasing and decreasing QTL alleles from 'Blenheim' were detected in more than one environment for all the sieving fractions. The most important for all 3 characters, in that it was detected the most frequently and produced the largest effects, was located in the region of *denso*, with the 'Blenheim' allele associated with a reduction in each fraction. Another QTL was detected on chromosome 3 in two environments for >22, in the region of *Rh*, and the 'Blenheim' allele also reduced the character. Two other QTLs were detected having effects on 2 of the 3 characters. One was located in two environments on chromosome 6 for >28 and in three environments for >25, with the 'Blenheim' allele increasing both. The other was detected in two environments on the short arm of chromosome 5 for >25 and >22, with 'Blenheim' alleles increasing the characters. Another QTL was located in two environments on chromosome 2 for >22 with the 'Blenheim' allele decreasing the character. Decreasing QTL alleles from

Table 2 Performance of parents and DH lines, percentage variation accounted for and numbers of effective factors (k) and of increasing (+) and decreasing (−) QTLs from 'Blenheim' detected for disease characters by regression

Character	Year[a]	Blenheim	E 224/3	DH Lines			k	%Variation	Blenheim alleles	
				Maximum	Mean[b]	Minimum			+	−[b]
1Ml	90	6.5	5.0	50.0	15.0	0.0	3.82	80.5	3	2
1Ml	92	27.5	8.8	50.0	19.7	0.6	4.34	37.0	2	0
2Ml	90	12.5	17.5	70.0	30.8	0.0	2.14	70.6	1	2
1BR	90	22.5	27.5	50.0	23.0	3.0	4.03	44.6	0	1
2BR	90	40.0	35.0	60.0	37.4	5.0	4.42	63.8	1	2
1YR	90	4.0	17.5	50.0	11.8	0.0	2.66	79.4	**3**	**2**
2YR	90	6.5	17.5	60.0	15.1	0.0	3.00	73.1	**2**	**1**
1Rh	90	3.0	0.5	30.0	5.5	0.0	4.34	68.2	3	2
1Rh	91	5.75	0.8	17.0	**6.3**	0.0	4.73	63.0	1	0
1Rh	92	17.25	0.0	25.0	10.1	0.0	3.19	72.5	3	0
2Rh	90	11.5	2.0	40.0	11.53	0.0	2.80	61.4	3	1
2Rh	91	23.0	0.5	75.0	**32.78**	0.05	3.45	77.4	3	1
2Rh	92	80.0	21.27	100.0	**72.46**	15.0	4.40	61.5	2	0

[a] Bold means significantly differ from mid-parent at 5% level
[b] Numbers in bold indicate significant interactions between QTLs

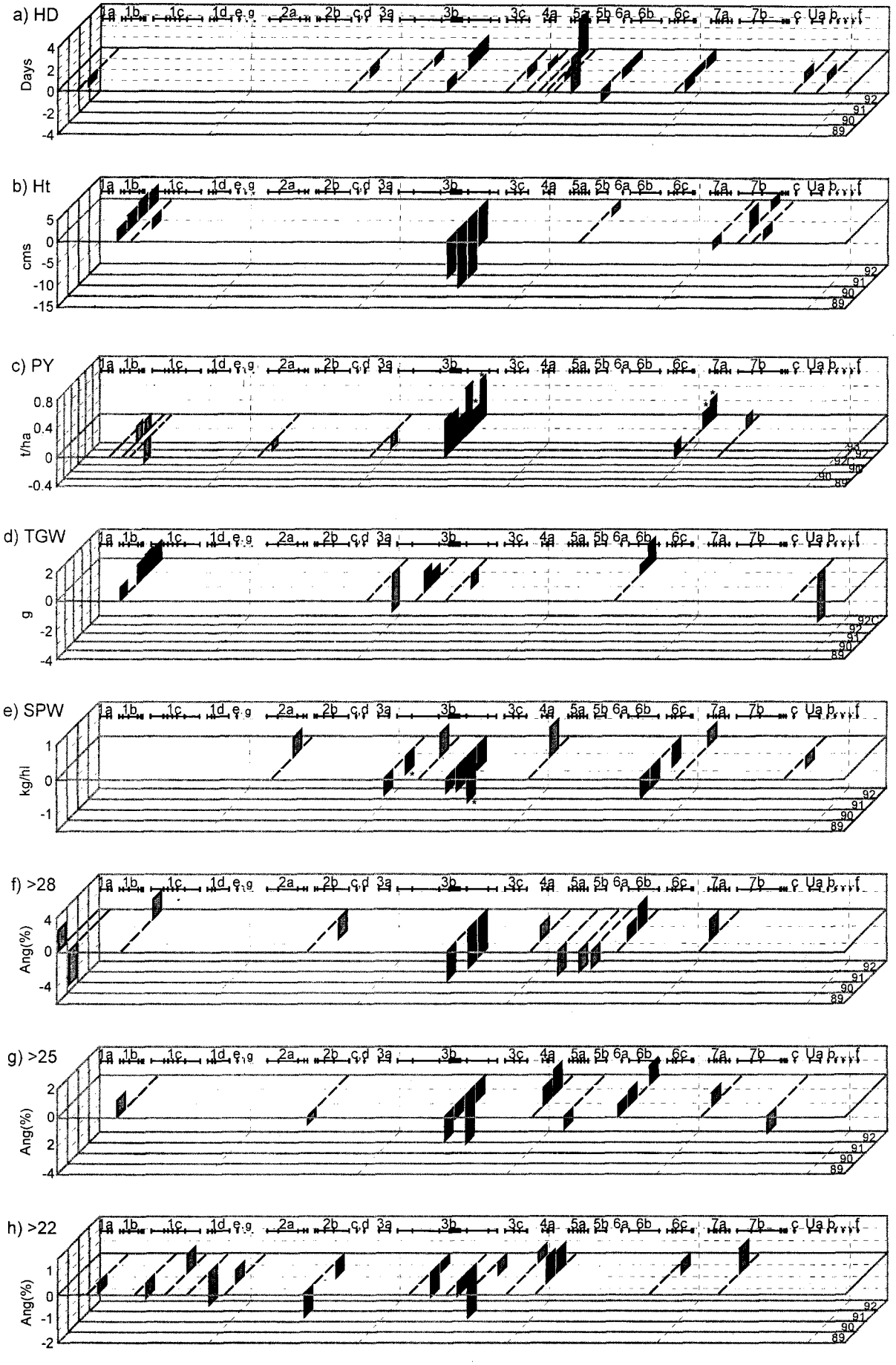
a) HD
Days
b) Ht
cms
c) PY
t/ha
d) TGW
g
e) SPW
kg/hl
f) >28
Ang(%)
g) >25
Ang(%)
h) >22
Ang(%)
1a 1b 1c 1d e g 2a 2b c d 3a 3b 3c 4a 5a 5b 6a 6b 6c 7a 7b c Ua b f
92
91
90
89

Fig. 2a–h Three-dimensional histograms showing location of QTLs for agronomic, yield and grain characters in each environment in which the character was scored. The deviation of *vertical bars* from the plane '0' on the Y-axis indicates the effect of the 'Blenheim' QTL allele. *Black bars* inidicate QTLs detected in more than one environment for a character and are located at the mean position over the environments in which they were detected. Interacting QTLs are indicated by *. The chromosome segments from Fig. 1 are arranged linearly at the *top* of the histograms. Results from PBIC scores are denoted by *C* after the year.

'Blenheim' were also found in the same region for >28 and >25, but only in one environment for each.

Disease resistance (1Ml, 2Ml, 1BR, 2BR, 1YR, 2YR, 1Rh and 2Rh) characters

Apart from 1Rh and 2Rh, the first and second scores for the disease characters were considered jointly. For mildew, the only QTL that was detected on more than one occasion was that in the region of *Hor1* and *Hor2* on chromosome 5, probably at the *Mla* locus, for 1Ml and 2Ml in 1990, with the 'Blenheim' allele increasing the area infected by the disease (Fig. 3a). A QTL allele from 'Blenheim' that reduced disease amount was detected for 1BR and 2BR on the long arm of chromosome 7 (Fig. 3b). Three QTLs were detected for both 1YR and 2YR, and these were located on chromosomes 1, 5 and 7. The QTL on chromosome 5 had the largest effect for both scores, with the 'Blenheim' allele reducing disease. 'Blenheim' alleles at the other 2 loci produced an increase in disease (Fig. 3c). Regression analysis revealed evidence of interaction between the QTL on chromosome 5 and those on chromosomes 1 and 7 for both 1YR and 2YR. This increased the portion of phenotypic variation ac-

Fig. 3a–e Three-dimensional histograms showing location of QTLs for disease characters in each environment in which the character was scored. The deviation of *vertical bars* from the plane '0' on the Y-axis indicates the effect of the 'Blenheim' QTL allele. *Black bars* indicate QTLs detected in more than one environment for a character and are located at the mean position over the environments in which they were detected. Interacting QTLs are indicated by *. The chromosome segments from Fig. 1 are arranged linearly at the *top* of the histograms

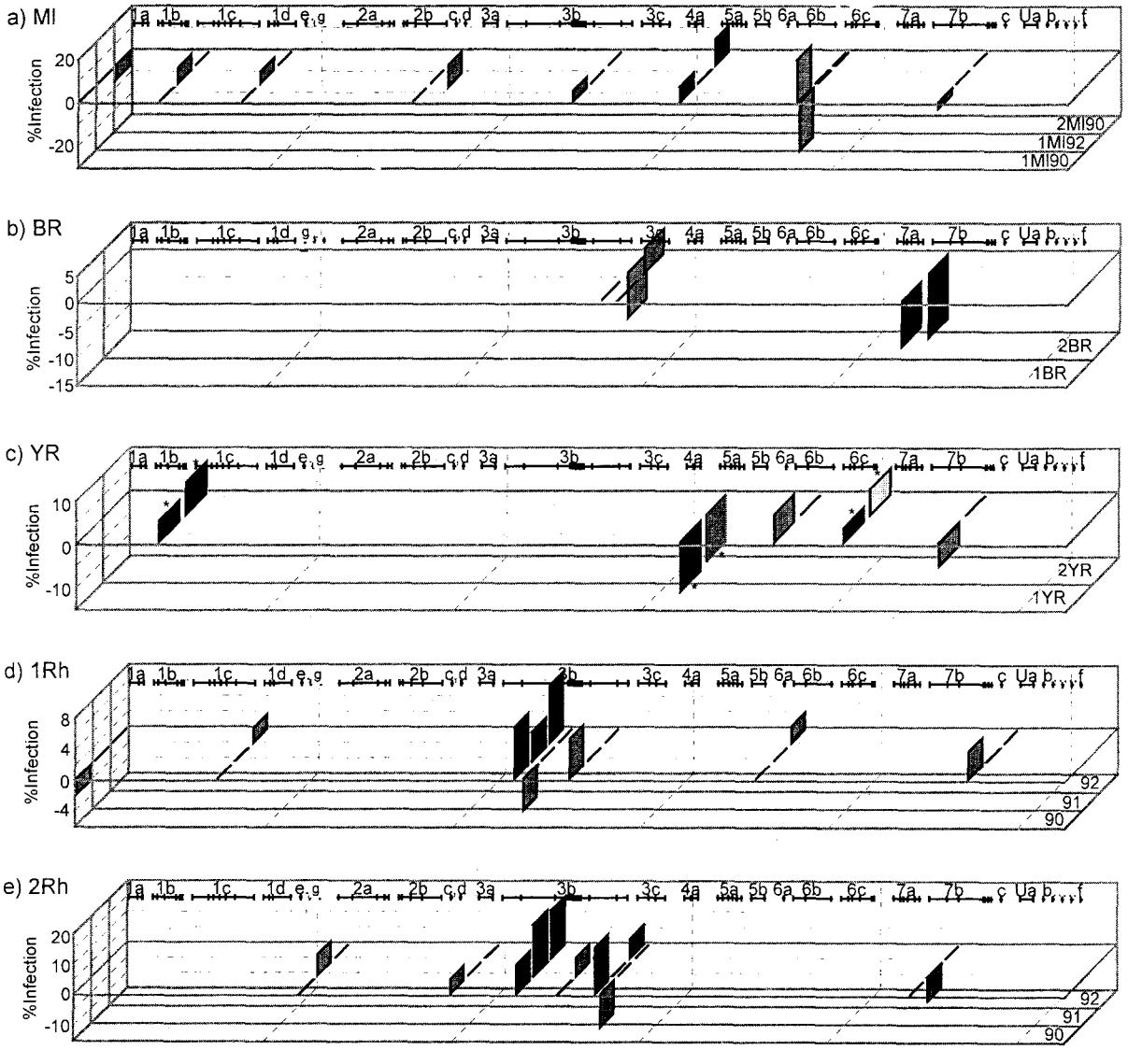

Table 3 Numbers of QTLs detected in up to six environments for each character

Character	Number of environments in which a QTL is present						Total QTLs in more than 1 environment
	6	5	4	3	2	1	
	Number of QTLs						
HD			0	3	0	10	3
Ht			2	0	1	4	3
PY	1	0	0	1	0	6	2
TGW		0	1	0	2	3	3
SPW			1	1	1	5	3
>28			0	1	1	9	2
>25			1	1	1	5	3
>22			0	0	4	9	4
Ml				0	1	8	1
BR					1	2	1
YR					3	2	3
1Rh				1	0	6	1
2Rh				1	1	5	2
Total	1	0	5	9	16	74	31

counted for to 87.2% for the former and 84.2% for the latter. For 1Rh and 2Rh, the *Rh* locus on chromosome 3 exerted a large effect over all three environments, with the 'Blenheim' alleles increasing disease, which is consistent with E 224/3 possessing an effective resistance gene. A second QTL for 2Rh was detected in 1990 and 1992 and was also located on chromosome 3, in the region of *Est 1,2* (Fig. 3e), with the 'Blenheim' allele also increasing disease.

We detected 159 QTLs over all the scores for the 13 characters under consideration in this paper (Table 3). Eleven QTLs were detected in each environment that the character was measured, 9 in all but one and 8 in all but two environments. A QTL for PY was detected in three out of the six environments, and 2 QTLs for TGW were detected in two out of the five environments in which the characters were measured, making 31 QTLs that were detected in more than one environment for the 13 characters.

Discussion

The cross in the present study was chosen from the SCRI breeding programme as it represented material typical of commercial North-West European spring barleys. It is interesting to compare our results with the barley QTL maps of Hayes et al. (1993b), Han and Ullrich (1994), Tinker et al. (1994) and Backes et al. (1995). The first two studies examined a fairly diverse cross in that one parent was a 6-row feed barley and the other a 6-row malting barley. The third was less diverse in that both parents were 2-row but still represented contrasting types for malting quality, agronomic characters and disease resistance. These three studies were conducted in North American spring barley germ plasm and the traits measured in North American environments, which differ considerably from their North-West European counterparts. The fourth study was of a cross between two North-West European 2-row winter barley cultivars, but there are considerable differences between the winter and spring barley gene-pools (Fischbeck 1991). In addition, none of the parents in the previous studies carried the *denso* dwarfing gene, which is carried by many of the widely grown North-West European spring barley cultivars.

It is obvious from the present study that the *denso* locus affects many of the characters. Comparison between maps suggests that the region around *denso* falls between the two mapped segments of chromosome 3 studied by Tinker et al. (1994) so that we cannot compare results from this region. Comparison with the map of Kleinhofs et al. (1994) suggests that the *denso* locus falls in the interval His4b-ABG4 studied by Hayes et al. (1993b) and Han and Ullrich (1994). In addition, it was not possible to align chromosome segments 1c and 5b from the present study with the other crosses due to the absence of any comparable markers. Six of the characters measured in the present study (HD, Ht, TGW, SPW and 1M1) are comparable to those of Hayes et al. (1993b), Han and Ullrich (1994) and Tinker et al. (1994). Whilst >25 is not exactly the same sieve size as that used by Han and Ullrich (1994) and Tinker et al. (1994), it is the nearest used in the present study and can be compared to the plump grains character used in the North American studies. Thirteen QTLs from the present study appear to be in the same region (i.e. within 25 cM) as some of those from the North American studies, and their approximate locations are listed in Table 4. Of these 13 QTLs 8 were detected in more than one environment but, in the present study, we detected 18 QTLs for the 7 common characters in more than one environment. Whilst there is therefore evidence of some similarities in the results, there is also evidence of considerable diversity between North American and North-West European spring barley germ plasm in QTLs

Table 4 QTLs in similar regions of the barley genome. Bold type indicates QTLs from 'Blenheim' × E224/3 detected in more than one environment

Chromosome	Character	Location		
		Blenheim × E224/3	Harrington × TR306[a]	Steptoe × Morex[b,c]
2	PY	OPW18–H700:CDO64a	-	Rbcs:ABG2
3	HD	ABG377	-	ABG396:ABG703A
3	HD	PSR540:Rh	-	ABG453:ABC307B
3	**TGW**	**OPO16-H900:SC101–24H500**	-	**ABG453:CDO113B**
3	**>25**	**WG940:denso**	-	**ABG453:CDO113B**
3	**PY**	**denso**	-	**CDO113B:His4B**
4	HD	PBI48	-	BCD402B:TubA1
6	**TGW**	**OPE11-H400:WG282**	**ABG705B**	**ksuD17:ABG474**
6	**>25**	**WG282**	**ABG705B**	-
6	**SPW**	**Amy1:PSR154**	**MWG934**	**Not published**
7	**HD**	**Rrn2:OPJ5-H850**	**CDO348:mSrh**	-
7	**PY**	**OPJ5-H850:OPE11-H1500**	**MWG502:ABG705A**	-
7	Ht	OPA19-H1000:CDO504	mSrh:ABG702B	CDO57B:mSrh

[a] Tinker et al. (1994)
[b] Hayes et al. (1993b)
[c] Han and Ullrich (1994)

controlling agronomic, yield and grain characters. Comparison with the study of Backes et al. (1995) is even more difficult, but where it was possible to align segments, there was no correspondence between any of the QTLs located by MAPMAKER/QTL in it and those located by regression analysis in the present study, probably reflecting the diversity between the winter and spring barley gene-pools noted by Fischbeck (1991).

Previous studies have not been able to discern whether the association of the *denso* dwarfing gene with characters other than height is due to linkage or pleiotropy. The fact that QTLs for PY and >25 were located in the region of *denso* in the present study and in a similar region in the cross studied by Hayes et al (1993b) and Han and Ullrich (1994), a cross in which neither parent possessed *denso*, could indicate that the association of *denso* with these 2 characters was due to linkage. Further evidence that *denso* might be linked to the QTL for PY is provided by the fact that the present study shows that the dwarf phenotype is associated with an increase in PY. There are a number of unrelated sources of the *denso* dwarfing gene, all of which have been shown to be allelic (Haahr and von Wettstein 1976). However, studies of the gene derived 'Abed Denso' (Snape and Simpson 1981b; Powell et al. 1985; Thomas et al. 1991) and of the 'Jotun' dwarf (Ali et al. 1978) revealed an association with a decrease in yield. Our study has examined the dwarfing gene derived from 'Diamant', which Gymer (1991) also found to be associated with yield increase. This suggests that the gene is probably very tightly linked to a QTL controlling PY and that the associations of the different sources of the gene depend upon the genetic background of their parents.

A number of genes for earliness (*Ea*) have been found in barley (Sogaard and von Wettstein-Knowles 1987). *Ea* genes generally have a much larger effect on HD than the QTLs detected in the present study, and those on the same chromosomes as the QTLs detected in more than one environment are located on opposite arms. The QTLs for Ht on chromosomes 1 and 7 probably represent the action of minor genes as their effect is small compared to that of the *denso* locus. The *erectoides* dwarfing genes are also used by barley breeders to reduce height; some have a large effects on Ht while others have a small effect. *Erectoides* genes are manifested by a reduction in rachis internode length (Persson and Hagberg 1969). Ears of some DH lines were quite compact but their rachis internode lengths were much greater than those typical of *erectoides* types. As no QTLs for rachis internode length were found at or near the QTLs for Ht (data not shown), it is unlikely that the QTLs on chromosomes 1 and 7 represent the action of *erectoides* genes.

The QTL from E224/3 on chromosome 5 that decreases mildew infection probably reflects the action of the *Mla13* mildew resistance gene possessed by the line (unpublished data) as the *Mla* locus is located between *Hor1* and *Hor2* (Sogaard and von Wettstein-Knowles 1987). This QTL was not detected in 1992, probably because of widespread virulence to *Mla13* in the barley mildew population at SCRI. There was no correspondence between the QTLs detected in the present study and those detected by Tinker et al. (1994), reflecting the different mildew resisitances and races of the pathogen found in North America. The *Yr4* Yellow Rust resistance locus is also located on the short arm of chromosome 5, close to *Hor1* (Sogaard and von Wettstein Knowles 1987). 'Deba Abed', an ancestor of 'Blenheim', carries the *Yr4* resistance (Stubbs 1985) and so the resistant Yellow Rust QTL from 'Blenheim' on chromosome 5 may be due to its effect. The other resistant Yellow Rust QTLs were derived from E224/3 and located on chromosomes 1 and 7 and, as both exert a considerable effect, could reflect the action of major genes. Chen et al. (1994) report a QTL for resistance to race 24 of Yellow Rust on chromosome 7, and as the

QTL we detected was in a similar region, it could therefore represent the action of this resistance. 'Triumph', a parent of 'Blenheim', possesses a seedling resistance to yellow rust, but this resistance may not be effective at adult plant growth stages (Meadway and Hutton 1994). The disease nursery measurements were all made on adult plants so none of the QTLs should represent the effect of the seedling resistance. 'Triumph' carries as well the *Rph12* resistance to Brown Rust, which is located on chromosome 7 (Jin et al. 1993), close to the region carrying the Brown Rust resistance QTL from 'Blenheim'. 'Blenheim' shares a common resistance with 'Triumph' (Jones and Clifford 1994) but was relatively susceptible to the 'Triumph' virulent race of Brown Rust prevalent in the disease nursery. If *Rph12* was the common gene, the QTL detected in the present study may represent its action in conjunction with 1 or more QTLs of minor effect from E224/3 which, individually, were too small to be detected in the present study.

In the present study, the large effect of a QTL was found to mask other QTLs controlling a character. For instance, a QTL at *denso* accounted for over 70% of the phenotypic variation in Ht and, no other QTLs were apparent until the *denso* QTL was fixed and the residuals examined by stepwise regression. Similarly, the QTL for N at *denso* accounted for a large portion of the variation in the character, and other probable QTLs were only revealed when it was fixed. The false location of a QTL may be a result (Haley and Knott 1992; Martinez and Curnow 1992), but the strategy we adopted was more valuable than a single scan as it revealed more probable QTLs. MAPMAKER/QTL can be used in stepwise strategy (Lander and Botstein 1989), but the technique used in this study has the advantage of incorporating unlinked markers. However, the differences between the current study and those of North American spring barley germ plasm (Hayes et al. 1993b; Han and Ullrich 1994; Tiner et al. 1994) and of winter barley germ plasm (Backes et al. 1995) emphasise the need for mapping to be conducted in crosses between genotypes of economic worth in their target locality for the results to have application in barley breeding.

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