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Linkage mapping of genes controlling endosperm storage proteins in wheat*

2. Genes on the long arms of group 1 chromosomes

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Summary. A translocation mapping procedure was used to map the *Glu-1* genes, controlling high-molecular-weight (HMW) glutenin subunits on the long arms of group 1 wheat chromosomes, with respect to their centromeres. The *Glu-A1* genes on chromosome arm 1AL were linked with the centromere but gave very different map distances (in cM) of 10.2 ± 3.5 and 33.9 ± 5.4 with test-cross and F_2 progeny, respectively. The centromere-*Glu-B1* and centromere-*Glu-D1* map distances were estimated to be 28.1 ± 2.8 and 30.9 ± 2.7, respectively, using test-cross seeds. A telocentric mapping procedure gave a reduced map distance of 22 ± 3.5 for the centromere-*Glu-D1* interval. Possible reasons for the differences between the mapping results obtained with test-cross and F_2 seeds and with the translocation and telocentric procedures are discussed.

Key words: Wheat – HMW glutenin subunits – Linkage mapping

Introduction

The long arms of chromosomes 1A, 1B and 1D of wheat carry the genes coding for HMW glutenin subunits (Orth and Bushuk 1974; Bietz et al. 1975; Brown et al. 1979, 1981; Lawrence and Shepherd 1980, 1981a; Payne et al. 1980, 1981; Galili and Feldman 1983a, b) and the loci have been designated as *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively (Payne et al. 1982). Two of these loci (*Glu-B1* and *Glu-D1*) are complex because *Glu-D1* always codes for two protein subunits and *Glu-B1* usually does also. The subunit pairs controlled by these two loci have been referred to as 1Dx/1Dy and 1Bx/1By, repectively (Payne et al. 1981), and the genes controlling the x and y subunits at each of these two loci are very tightly linked (Payne et al. 1980; Lawrence and Shepherd 1981b). Recently, Payne et al. (1984) observed a rare recombination within the *Glu-B1* locus, but no such recombination has been reported within the *Glu-D1* locus.

Using the telocentric mapping procedure of Sears (1962, 1966), Payne et al. (1982) showed that the *Glu-A1, Glu-B1* and *Glu-D1* genes recombine with their respective centromeres with frequencies of 7.6%, 9.2% and 10.1%, respectively. However, these recombination values may underestimate the true values since it has been shown in cotton (Endrizzi and Kohel 1966) and wheat (Sears 1972) that loci near the centromere show much less recombination in telocentric mapping compared to mapping with the complete chromosome.

In the present study the translocation method for mapping gene-centromere distances (Singh and Shepherd 1984, 1988) was used to map the position of the *Glu-1* loci on the long arm of each of the group 1 wheat chromosomes. These results are compared with the telocentric mapping results obtained from a telocentric mapping experiment in the present study and also with those obtained by Payne et al. (1982).

Materials and methods

1 Genetic stocks

Wheat cultivars Gabo, Halberd, Heron, Kite and India 115; 'Single glutenin line' which lacks the HMW glutenin subunits controlled by chromosomes 1A and 1D (Lawrence and Shepherd 1981b); and Chinese Spring (CS) ditelocentric (Dt) lines Dt 1AL and Dt 1DL (Sears 1954; Sears and Sears 1978) are all maintained at the Waite Agricultural Research Institute. Seeds of the breeding lines MKR/211/7 and WR/17 were kindly provided by Dr. A. J. Rathjen of this Institute.

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The 1DL-1RS translocation line was isolated by Shepherd (1973) from among the progeny of a CS wheat plant double monosomic for chromosomes 1D and 1R ($20^{II}+2^{I}$). The chromosome arm 1RS was derived from the CS-Imperial rye addition line 'E' produced by Driscoll and Sears (1971). This translocation has been backcrossed into many Australian wheats and two of these namely Warigal 1DL-1RS and Halberd 1DL-1RS were used in the present study. The 1BL-1RS translocation was isolated by Dr. K. W. Shepherd (unpublished) while attempting to transfer the complete 1R substitution of Imperial rye for 1B in CS to a Gabo background by backcrossing. The translocation was isolated during the 5th backcross to Gabo. The 1AL-1RS translocation was produced in a CS background during the present study, using the method of Shepherd (1973). However, since chromosome arm 1AL of CS does not carry any seed storage protein marker, the double monosomic line required as starting material, was produced by crossing CS-Hope 1A substitution line (Sears et al. 1957) to a CS-Imperial rye 1R(1A) substitution line. Thus the chromosome arm 1AL in this translocation is derived from the cultivar Hope. Cytological and biochemical studies suggest that each of these three translocations resulted from the centromeric fusion of freshly produced wheat and rye telocentrics due to misdivision of univalents in the double monosomics.

2 Gene mapping procedures

The principle of the translocation mapping procedures used to map the *Glu-1*-centromere distances has been described before (Singh and Shepherd 1984, 1988) and it assumes that the translocations used involved centromeric fusion between 1RS and the long arm of each group 1 chromosome. The parents were chosen so that in the F_1 each of the four arms of the bivalent involving the translocation had recognizably different endosperm protein markers. The F_1 heterozygotes were test-crossed as female to a third parent which ideally had no endosperm protein bands overlapping those to be scored in the progeny. The secalin protein markers on 1RS and gliadin markers on 1AS, 1BS or 1DS were used to mark the position of the centromere. A telocentric mapping experiment (Sears 1962, 1966) with the *Glu-D1* locus was carried out in parallel with the translocation mapping in order to compare the map distances obtained with these two methods.

A summary of the test-cross combinations used for mapping the *Glu-A1*, *Glu-B1* and *Glu-D1* loci employing translocation chromosomes and for mapping the *Glu-D1* locus using a telocentric chromosome, is given in Fig. 1. The various gene and allele symbols have been described in detail earlier (Singh and Shepherd 1988).

3 Cytological analysis

Pollen mother cells (PMCs) from the F_1 heterozygotes were analysed to find the degree of pairing between the wheat-rye translocation chromosomes or 1DL telocentric and their normal group 1 wheat chromosome homologues. Anthers at metaphase I of meiosis were fixed in three absolute ethanol : 1 glacial acetic acid for 12–24 h at 4 °C, hydrolysed in 1N HCl at 60 °C for 14 min and then treated with Feulgen reagent for approximately 2 h at room temperature before being squashed in 45% acetic acid for microscopic examination.

4 Extraction and electrophoretic separation of seed proteins

Seed proteins were extracted from the endosperm halves of the single kernels as described earlier (Singh and Shepherd 1988). However, in some experiments better resolution of

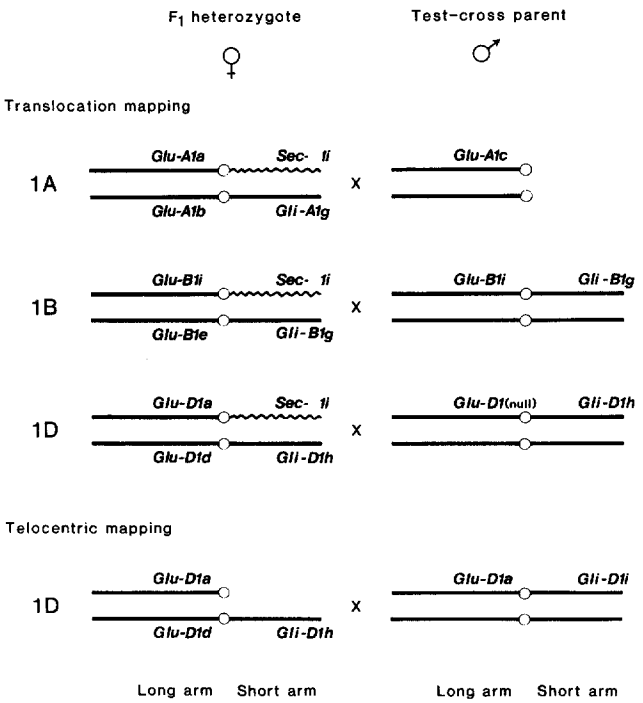

Fig. 1. Test-cross combinations used for mapping genes coding for wheat endosperm proteins on the long arms of group 1 chromosomes. Translocation mapping: *1A* (CS-Hope 1AL-1RS × Gabo) × CS Dt 1AL; *1B* (Gabo 1BL-1RS × MKR/211/7) × Single glutenin line; *1D* (CS 1DL-1RS × Halberd) × Single glutenin line. Telocentric mapping: *1D* (CS Dt 1DL × Halberd) × India 115. *CS* = Chinese Spring. The gene and allele symbols are explained in Table 1 of Singh and Shepherd (1988)

prolamins was obtained using an ethanol [30% (v/v)]-sucrose [20% (w/v)] mixture as the extractant at 25 °C. The sample buffer (pH 6.8) containing 2-mercaptoethanol was added to the same tubes for the subsequent extraction of glutenins. One-dimensional sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) was used as described earlier (Singh and Shepherd 1988).

5 Genetic analysis

In the test-cross mapping, estimation of recombination frequencies with standard errors and their conversion into map distance (cM) was based on methods of Mather (1951) and Kosambi (1944) as described earlier (Singh and Shepherd 1988).

With F_2 mapping, the recombination frequency and its standard error was calculated using the maximum likelihood method (Haldane 1919; Mather 1951). Since endosperm protein phenotypes are codominant, the F_2 progeny could be classified into 9 out of 10 possible genotypic classes in a completely classified F_2 progeny segregating for two factors. The two classes that could not be distinguished comprised those individuals having all four alleles of the two loci being mapped, representing seeds with either both parental or both recombinant type chromosomes. Assuming equal recombination frequencies in male and female gametes, the maximum likelihood method was used to derive the following cubic

equation for calculating recombination fraction (p) from a nine class F_2 data.

$$(b+2d)-2p(a+2b+c+3d)+2p^2(2a+3b+3c+4d)-4p^3(a+b+c+d)=0$$

where,

a = combined observed frequency of the two homozygous parental classes;
b = combined observed frequency of the four classes involving one parental and one recombinant gamete;
c = observed frequency of the double heterozygote class showing all four alleles;
d = combined observed frequency of the two homozygous recombinant classes.

The equation used to calculate the standard error (S_p) of the recombination fraction was also derived using maximum likelihood method (Mather 1951) as follows:

$$\frac{1}{(S_p)^2} = 2n\left[1+\frac{(1-2p+2p^2)}{p(1-p)}-\frac{2(1-3p-3p^2)}{(1-2p+2p^2)}\right]$$

Results

1 Cytological analysis of the F_1 heterozygotes

Unlike the tanslocation chromosomes involving chromosome arm 1RL of Imperial rye (Singh and Shepherd 1988), the 1RS translocations could not be recognized in C-banded preparations and, therefore, Feulgen preparations were used.

The 1AL-1RS and 1A chromosomes were not easily identifiable and therefore, the two chromosomes were considered to be definitely paired in only those PMCs showing 21 bivalents. This practice will lead to an underestimation of pairing because in some cells the univalents may be chromosomes other than 1A or 1AL-1RS. As evident from the lack of aneuploids amongst the test-cross progeny (Table 2), pairing between these two chromosomes must have been very high and the observed 89.5% pairing (Table 1) is most probably an underestimate.

The 1BL-1RS and 1B chromosomes could be identified by the presence of a satellite on 1BS and slight heteromorphy of the bivalent involving these chromosomes. A total of 303 PMCs were examined and 96.4% showed 21 bivalents (Table 1); therefore asynapsis was insignificant and it is reflected in the low number of aneuploids in the test-cross progeny (Table 4).

The translocation chromosome 1DL-1RS was slightly larger than chromosome 1D and these two were easily identified by their heteromorphy in the Feulgen preparations of PMCs at metaphase I. The bivalents involving 1DL telocentric were also clearly heteromorphic. As shown in Table 1, the complete 1D chromosome paired more frequently with the 1DL-1RS tanslocation (90.7%) than with the 1DL telocentric (83.6%).

A total of 644 PMCs from the three different translocation heterozygotes were inspected and there was no evidence of rye chromosome arm 1RS pairing with any of the wheat chromosomes, suggesting that all these translocations arose from centric fusion.

Table 1. Observed frequencies of pairing between complete wheat chromosomes and the homologous long arms of wheat-rye translocations and the 1DL telocentric chromosomes at metaphase I

F_1 combination	Season grown	No. of PMCs	No. paired	% pairing
CS-Hope 1AL-1RS × Gabo	Spring, 1983	190	170[a]	89.5
Gabo 1BL-1RS × MKR/211/7	Spring, 1982	303	292	96.4
CS 1DL-1RS × Halberd	Spring, 1982	151	137	90.7
CS Dt 1DL × Halberd	Spring, 1982	128	107	83.6

[a] Cells with 21 bivalents, 1A/1AL-1RS bivalent not positively identified

2 Gene mapping

2.1 Mapping the Glu-A1 *locus on chromosome arm 1AL.* The HMW glutenin subunits coded by *Glu-A1a* and *Glu-A1b* and the wheat and rye prolamins coded by *Gli-A1g* and *Sec-li* were classified without difficulty in the parents (Fig. 2 A, B, a, b). Chromosome arm 1AS

Table 2. Observed frequencies of parental and recombinant protein phenotypes in test-cross progeny used to map the *Glu-A1* locus

Protein phenotypes				Total	Recombination (%)	Map distance (in cM ± SE)
Parentals		*Recombinants*				
Glu-A1a Sec-1i	Glu-A1b Gli-A1g[a]	Glu-A1a Gli-A1g[a]	Glu-A1b Sec-1i			
31	40	3	5	79	10.1	10.2 ± 3.5

[a] Tri-A1 was also present

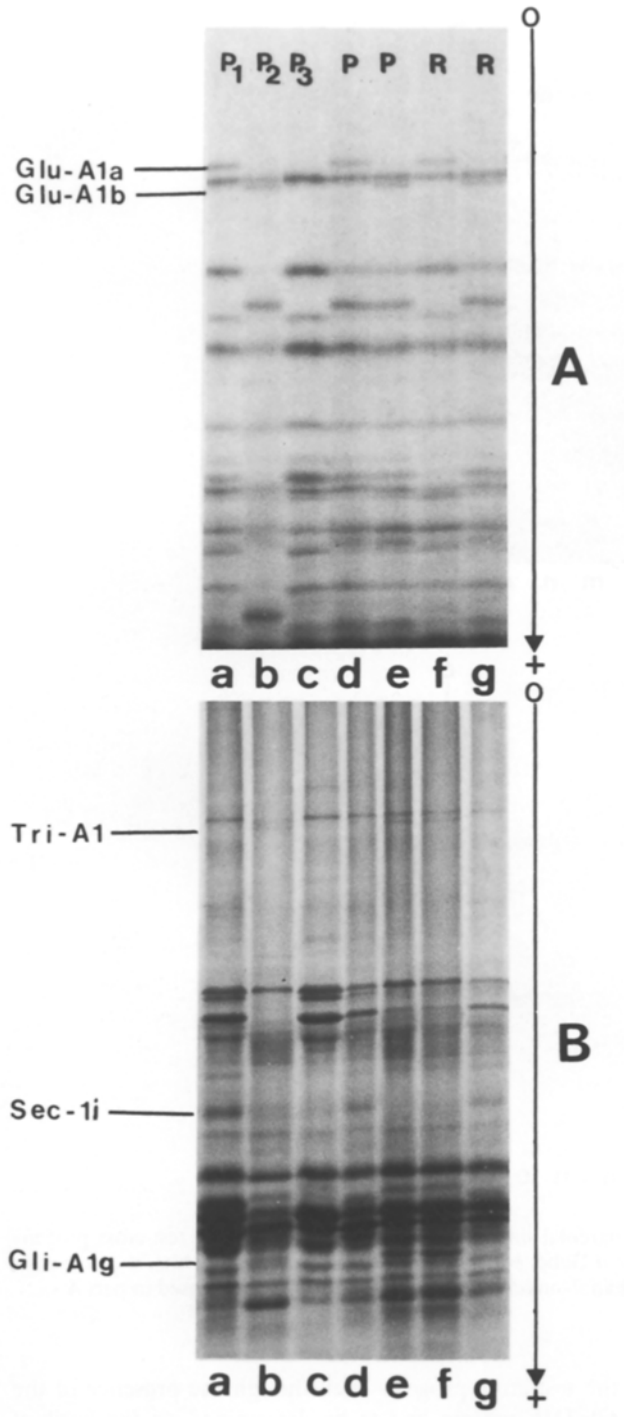

Fig. 2 A, B. SDS-PAGE patterns of total seed protein extracts from the parental lines (P_1, P_2, P_3) and some of the test-cross progeny used to map the *Glu-A1* locus. **A** reduced proteins: P_1 CS-Hope 1AL-1RS, P_2 Gabo, P_3 CS Dt 1AL, *d–g* test-cross seeds, *P* = parental, *R* = recombinant; **B** unreduced proteins obtained from the same seeds used in part **A**

also controls a protein Tri-A1 which results in a hybrid triplet band (Fig. 2 B, b; Singh and Shepherd 1985, 1988). Since the male test-cross parent CS Dt 1AL lacked all of these bands the test-cross progeny could be unambiguously classified into parental and recombinant types (Fig. 2 A, B, d–g). No aneuploid was detected among these progeny. A total of 79 test-cross seeds were analysed, and since the data were homogeneous ($\chi^2_{[3]} = 2.67$, $P > 0.2$), pooled values are given in Table 2. Single factor segregations for *Glu-A1a/Glu-A1b* and *Gli-A1g/Sec-1i* agreed with the expected 1 : 1 ratio ($\chi^2_{[1]} = 1.53$ and 0.63, respectively). Linkage was detected between *Glu-A1* and the centromere ($\chi^2_{[1]} = 50.2$) and the recombination frequency was 10.1%.

In addition, 158 F_2 seeds from 3 different F_1 plants were analysed and because of codominance these progeny could be classified into nine phenotypic classes. These F_2 data are more efficient than test-cross data for the measurement of linkage and are equal to test-cross data for the detection of linkage (Mather 1936). The data from the three different plants were homogeneous ($\chi^2_{[6]} = 9.76$, $P > 0.1$) after grouping into four phenotypic classes, and they are pooled in Table 3. Once again *Glu-A1a/Glu-A1b* ($\chi^2_{[1]} = 0.76$) and *Gli-A1g/Sec-1i* ($\chi^2_{[1]} = 1.30$) followed the expected 1 : 1 segregation ratio and the linkage between *Glu-A1* and the centromere was significant ($\chi^2_{[1]} = 24.88$). The recombination frequency was calculated to be 29.5% using the maximum likelihood method, and it is almost three times that obtained with the test-cross seeds (10.1%).

2.2 *Mapping the* Glu-B1 *locus on chromosome arm 1BL.*

Three different test-cross combinations were used to produce a total of 430 seeds in summer and spring of 1982. The HMW glutenin subunits coded by *Glu-B1i* and *Glu-B1e* and the prolamins coded by *Gli-B1g* and *Sec-1i* could be easily classified in the parents (Fig. 3 A, B, a, b). The HMW glutenin subunits and the prolamins were scored in the gels after SDS-PAGE of the reduced and unreduced proteins, respectively. Among the test-cross progeny, Glu-B1e and Sec-1i were classified without difficulty, but Glu-B1i and Gli-B1g proteins were present in the male parent (P_3) also, and they could only be scored by differences in staining intensity. However, this classification could be cross-checked because these two protein bands segregated as alternatives to the easily identifiable bands Glu-B1e and Sec-1i, respectively (Fig. 3 A, B, tracks d-p). Data obtained from the three test-cross families were homogeneous ($\chi^2_{[8]} = 8.55$, $P > 0.3$) and they are pooled in Table 4. The *Glu-B1i/Glu-B1e* and *Gli-B1g/Sec-1i* segregation agreed with the expected 1 : 1 ratio ($\chi^2_{[1]} =$ 0.39 and 1.23, respectively). However, linkage was detected ($\chi^2_{[1]} = 102.3$) between the centromere and

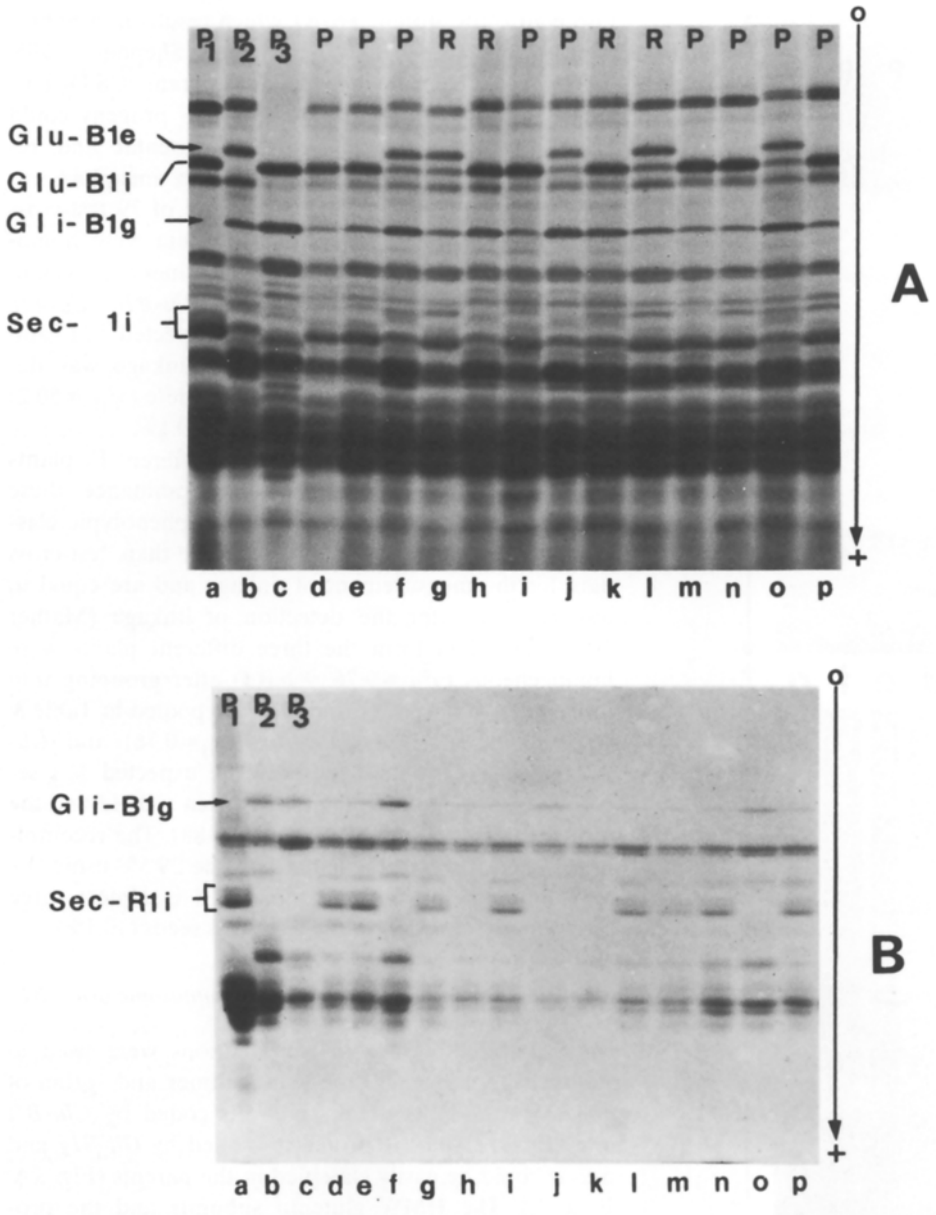

Fig. 3 A, B. SDS-PAGE patterns of seed protein extracts from the parental lines (P_1, P_2, P_3) and some of the test-cross progeny used to map the *Glu-B1* locus. **A** reduced total seed protein extracts: *a* Gabo, *b* (MKR) 211/7, *c* Single glutenin line, *d–p* test-cross progeny, *P* = parental, *R* = recombinant; **B** unreduced prolamins (ethanol-sucrose extracts) from the same seeds used in part **A**

Glu-B1 locus, with a mean recombination frequency of 25.5%.

2.3 Mapping the Glu-D1 *locus on chromosome arm 1DL.* Four test-cross combinations for translocation mapping and two for telocentric mapping were used to produce 529 and 191 seeds, respectively. The test-cross seeds were produced in summer and spring of 1982. The HMW glutenin subunits coded by *Glu-D1a* and *Glu-D1d* and the prolamins coded by *Gli-D1h* and *Sec-1i* were easily classified in the parents (Fig. 4 b, c) and the test-cross progeny even though the presence of the Gli-D1h protein had to be determined on the basis of band staining intensity (Fig. 4 e-k) because of overlap with the Gli-D1 band from the male parent. The scoring of Gli-D1h and Sec-1i bands could be cross-checked by SDS-PAGE of unreduced proteins similar to the scoring of Gli-B1g and Sec-1i bands above (Fig. 3 B).

The data obtained from the six test-crosses, excluding the aneuploid frequency, were homogeneous ($\chi^2_{[15]} = 17.35$, $P > 0.2$). However, the translocation and

Table 3. Observed frequencies of protein phenotypes in F_2 progeny used to map the *Glu-A1* locus

Protein phenotypes				No. of progeny	Total	Expected frequency	Recomb. (%)	Map distance (in cM ± SE)
Glu-A1a	Glu-A1b	Sec-1i	Gli-A1g[a]					
+	–	+	–	16			29.5	33.9 ± 5.4
–	+	–	+	15	31	$0.5\,(1-p)^2$		
+	+	+	–	12				
+	+	–	+	27				
+	–	+	+	15				
–	+	+	+	8	62	$2p\,(1-p)$		
+	+	+	+	58	58	$0.5p^2+0.5\,(1-p)^2$		
+	–	–	+	5				
–	+	+	–	2	7	$0.5\,p^2$		
Total				158	158	1		

[a] Tri-A1 was also present; p = proportion of recombinant gametes; 1–p = proportion of parental gametes; '–' = protein absent; '+' = protein present

Table 4. Observed frequency of parental, recombinant and aneuploid protein phenotypes in test-cross progeny used to map the *Glu-B1* locus

Protein phenotypes				*Aneuploids*	Total	Recombination (%)[a]	Map distance (in cM ± SE)
Parentals		*Recombinants*					
Glu-B1i Sec-1i	Glu-B1e Gli-B1g	Glu-B1i Gli-B1g	Glu-B1e Sec-1i				
150	168	57	52	3	430	25.5	28.1 ± 2.8

[a] Aneuploids excluded

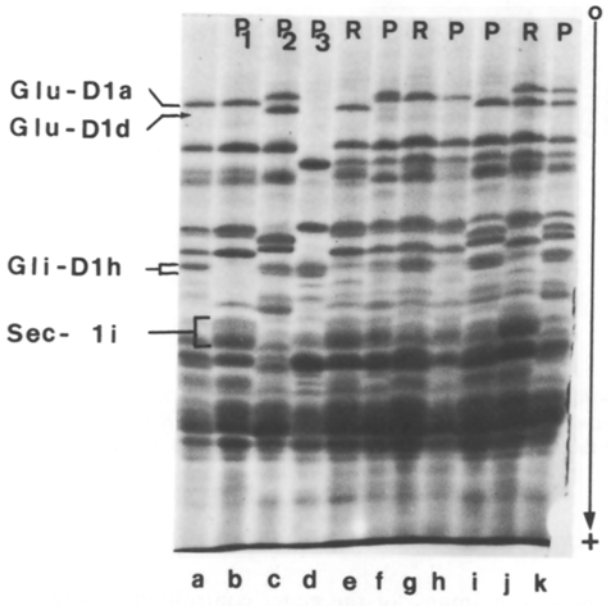

Fig. 4. SDS-PAGE patterns of reduced total seed protein extracts from the parental lines (P_1, P_2, P_3) and some of the test-cross progeny used to map the *Glu-D1* locus. *a* CS, *b* CS 1DL-1RS, *c* Halberd, *d* Single glutenin line, *e–k* test-cross progeny; *P* = parental, *R* = recombinant

telocentric mapping experiments are considered separately in Table 5 for comparison. With the translocation, the *Glu-D1a/Glu-D1d* and *Gli-D1h/Sec-1i* segregations followed the expected 1 : 1 ratio ($\chi^2_{[1]}=1.125$ and 0.07, respectively), but there was a significant linkage component ($\chi^2_{[1]}=103.3$). Similar conclusions were drawn from the χ^2 values for the single factor segregations and joint segregation in the telocentric mapping ($\chi^2_{[1]}=1.97$, 0.92 and 62.5, respectively). However, the recombination frequency measured with the translocation mapping (27.5%) was much higher than that obtained with the telocentric procedure (20.7%). As expected from the pairing data (Table 1), the proportion of aneuploid gametes was higher with the telocentric mapping (4.2%) than the translocation mapping (3.2%).

2.4 Recombination within the Glu-D1 *locus.* Two protein bands controlled by *Glu-D1a* (subunits 2 and 12 designated as 1Dx and 1Dy, respectively) and two alternative bands controlled by the allele *Glu-D1d* [subunits 5(1Dx) and 10(1Dy)] are indicated in Fig. 5. Each pair of bands is normally inherited as a unit and

Table 5. Observed frequency of parental, recombinant and aneuploid protein phenotypes in test-cross progeny used to map the *Glu-D1* locus

Protein phenotypes				*Aneuploids*	Total	Recombination (%)[b]	Map distance (in cM ± SE)
Parentals		*Recombinants*					
Glu-D1a (Sec-1i)[a]	Glu-D1d Gli-D1h	Glu-D1a Gli-D1h	Glu-D1d (Sec-1i)[a]				
Translocation mapping							
186	185	58	83	17	529	27.5	30.9 ± 2.7
Telocentric mapping							
71	74	11	27	8	191	20.7	22.0 ± 3.5

[a] Not present in the telocentric mapping; [b] Aneuploids excluded

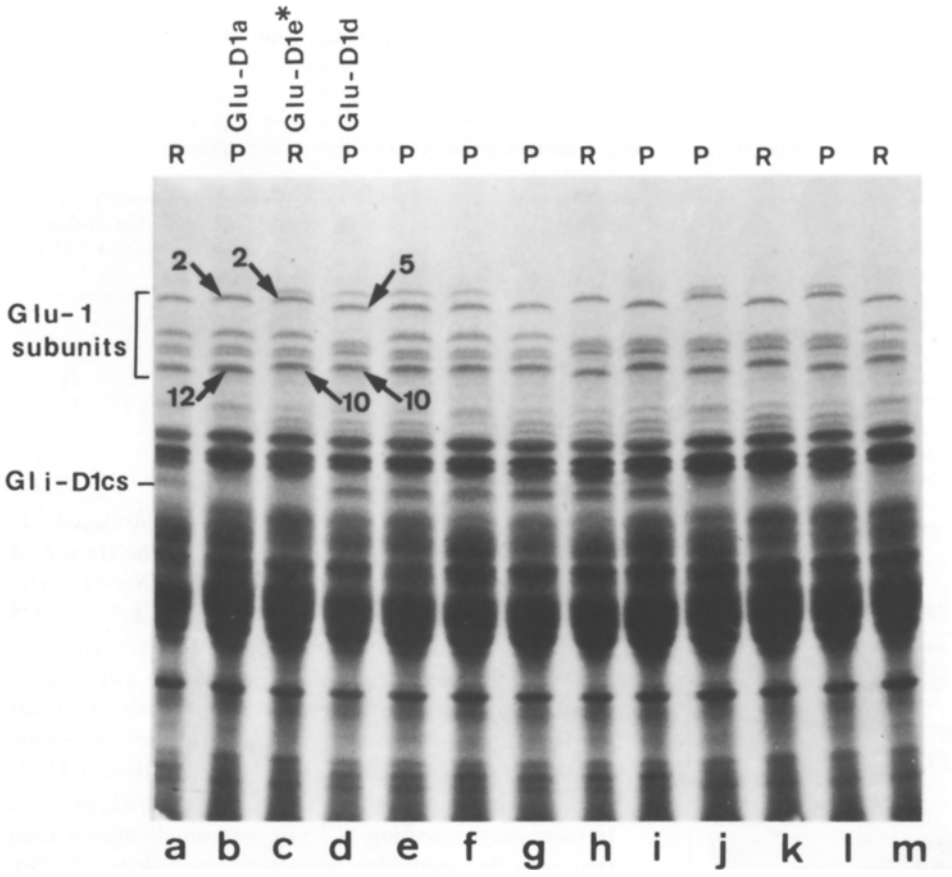

Fig. 5. SDS-PAGE patterns of reduced total protein extracts from some of the test-cross progeny used to map the *Glu-D1* locus. * Rare recombinant combining subunits 2 and 10 due to crossing over within the *Glu-D1* locus. *P* = Parental and *R* = Recombinant due to crossing over between centromere (marked with Gli-D1cs) and the *Glu-D1* locus

no recombinant type (2 + 10 or 5 + 12) was detected in 138 F_2 seeds analysed by Payne et al. (1980) and a further 479 test-cross seeds analysed by Lawrence and Shepherd (1981 b). However, in the present study a rare recombination between subunits 2 and 10 was observed (Fig. 5 c) among the 139 progeny from a test-cross (CS 1DL-1RS × Heron) × Single glutenin line.

3 Linkage maps

The linkage maps for the genes controlling endosperm storage proteins on the group 1 wheat chromosomes are shown in Fig. 6, and it also includes a summary of the data obtained in the preceding paper (Singh and Shepherd 1988).

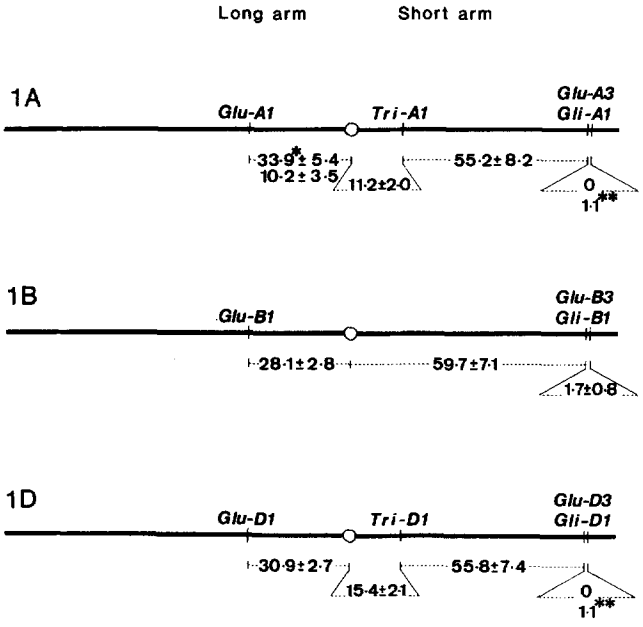

Fig. 6. Linkage maps of group 1 wheat chromosomes showing the location of genes coding for the endosperm storage proteins * F_2 mapping; ** upper limit

Discussion

The translocation method for mapping gene-centromere distance is based on the assumption that pairing does not occur between the homoeologous wheat and rye chromosome arms and that the homologous (wheat/wheat) arms of the translocations and complete wheat chromosomes pair normally. In fact, the observations made at metaphase I of meiosis did not show any indication of homoeologous pairing and there was a high level of pairing between the homologous long arms of the translocations and complete wheat chromosomes. Due to the low number of univalents at metaphase I, the number of aneuploids among test-cross progeny was also very low ($\leqq 3.2\%$). Similarly, pairing between the 1DL telocentric and the complete 1D chromosome was also high and as a result only a small proportion of the test-cross progeny were aneuploids (4.2%). Thus, it is concluded that the lack of chromosome pairing had very little effect on the estimates of map distances, particularly with the translocation mapping.

The *Glu-1* genes coding for the HMW glutenin subunits were found to be linked to the centromere, although two different map distances, 10.2 ± 3.5 cM from test-cross and 33.9 ± 4 cM from F_2 data, were obtained for the *Glu-A1*-centromere interval. Since the F_2 seeds were obtained from the same plants which were used to produce test-cross seeds, this difference could not be due to the effet of genetic background nor environment because both groups of seeds were being produced at the same time. It is possible however, that there was a much higher recombination frequency in the PMCs compared to the megaspore mother cells (MMCs) and further work is needed to resolve this question. The map distances between the *Glu-B1* and *Glu-D1* loci and their respective centromeres (28.1 ± 2.8 cM and 30.9 ± 2.7 cM, respectively) were comparable to the F_2 mapping results with the *Glu-A1* locus. However, these map distances are approximately three times higher than the average value (9.0 ± 1.2 cM) reported by Payne et al. (1982) for the same gene intervals using telocentric mapping procedures.

Telocentric mapping was also used in the present study to map the *Glu-D1* locus. Although the estimated map distance (22.7 ± 3.5 cM) was still much higher than the value (10.2 ± 2.4) obtained by Payne et al. (1982), it was significantly lower than that obtained with the translocation mapping (30.9 ± 2.7). The difference between the telocentric mapping results of Payne et al. (1982) and the current studies may be due to differences in the environmental conditions, particularly temperature, under which the plants were grown, or difference in the genetic backgrounds of the cultivars used.

However, the consistently lower map distance obtained with the telocentric mapping procedure compared to translocation mapping is most probably due to a reduction in the frequency of crossing over near the centromere when one member of the bivalent involved is a telocentric. For example, Endrizzi and Kohel (1966) found only 1.0% and 4.4% exchange with the centromere for two genes located on opposite arms of chromosome 6 of cotton, using the respective telocentrics, but obtained 22.1% recombination between the same genes when complete chromosomes were involved. Although the translocation chromosomes used in the present study possess alien chromosome segments from rye, nevertheless, they were complete chromosomes with two arms. In fact, Sears (1972) has shown in wheat that even where most of one chromosome arm consisted of alien chromatin, the recombination frequency in the region between the centromere and an awn inhibitor gene '*B*' was increased almost four fold compared to the value obtained with telocentric mapping (3.5% vs 0.87%). However, this reduction in recombination frequency observed with loci proximal to the centromere, was not significant for loci which were distal to the centromere (Endrizzi and Kohel 1966; Sears 1972). Also, Singh and Shepherd (1988) obtained similar results when telocentric and translocation mapping procedures were compared in mapping the distal *Gli-B1* locus with respect to the centromere (40.4% and 41.6%, respectively). The proximal loci studied by Endrizzi and Kohel (1966) and Sears (1972) were much closer to the centromere than the *Glu-D1* locus in the present study and this may explain why we obtained a less pronounced reduction in the recombination frequency with the telocentric mapping.

Although translocation mapping has consistently given higher map distances than telocentric mapping, further work is required to find whether the former values are similar to those occurring with complete homologues. The higher chromosome pairing frequencies observed between homologous arms of complete wheat chromosomes and wheat-rye translocations, compared to those observed between telocentrics and complete chromosomes (Singh and Shepherd 1988 and this paper), indicate that the rye chromosome arm did not interfere with the pairing of the other wheat arm. However, it is not known how these pairing frequencies compare with those occurring between pairs of complete homologous chromosomes.

The rare recombination between the 1Dx and 1Dy subunits of glutenin controlled by the *Glu-D1* locus may explain the origin of some of the naturally occurring rare patterns which combine x and y subunits from

two different frequently occurring alleles. For example, Payne and Lawrence (1983) consider that the French wheat cv. 'Flinor' has glutenin subunits 2+10 and they have designated this allele '*Glu-D1e*'. The recovery of this 'allele' from the parents having alleles *Glu-D1a* and *Glu-D1d* by recombination suggests that the two bands are the products of two very tightly linked genes. Recently, Branlard and Le Blanc (1985) concluded that the French cvs. Flinor and Elite possess the subunit combination 2+11 rather than 2+10. If they are correct then the 2+10 recombinant reported here represents a new combination of Glu-D1 bands.

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