Genetic control of the rate of transmission of rye B chromosomes. IV. Localization of the genes controlling B transmission rate

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Crosses between rye plants from selected lines for high and low B chromosome transmission rate were carried out to determine the location of the genes controlling B transmission rate. Our results show that they are located on the Bs and we hypothesize that such 'genes' are sites for chiasma formation. Our results also suggest that rye B chromosome polymorphism is controlled mainly by the Bs.

Keywords: B chromosomes, B transmission, rye, *Secale cereale*.

Introduction

Rye B chromosome transmission is non-Mendelian owing to the mechanism of accumulation and to the variable behaviour of the Bs during meiosis and gametogenesis (see Jones & Puertas, 1993 for a review). The mechanism of accumulation consists of nondisjunction at the first division of the male and female gametophytes and preferential segregation to the nucleus which will form the gametes.

B transmission rate in $2B \times 0B$ crosses is highly variable (Puertas *et al*., 1990). In some crosses the Bs tend to be lost showing a mean of less than 1 B per plant in the progeny, whereas others show B accumulation. In previous papers (Romera *et al*., 1991; Jiménez *et al*., 1995) we have shown that rye B transmission rate is genetically controlled. We selected genotypes of high (H) and low (L) mean number of Bs in the progeny of $2B \times 0B$ crosses. B loss in the L line is caused by a high frequency of B univalents at metaphase I which results in their loss during pollen grain development. The B accumulation in the H line results from regular meiotic behaviour followed by nondisjunction at first pollen metaphase (Jiménez *et al*., 1997).

In the present paper we show the results of crosses designed to determine if the rye B transmission control genes selected in the L and H lines are located on the normal chromosome set (As) or on the Bs. The implications for understanding B polymorphism in natural populations are different in

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each case. To date, the situation has been seen as a 'coevolutionary race' between the mechanism of accumulation of the Bs and genotypes on the As which may evolve counteracting the accumulation of the Bs (Beukeboom, 1994; Camacho *et al*., 1996), or looked upon as As and Bs in some species having a host–parasite interaction with coevolutionary forces acting to determine equilibrium frequencies in populations (Jones, 1995).

Materials and methods

The selection process of the L and H lines for the character *mean number of Bs per plant in the progeny* was described in Romera *et al*. (1991) and Jiménez *et al*. (1995). In summary it consisted of selecting plants from $2B \times 0B$ crosses of the Puyo population showing a strong tendency to lose or to accumulate the Bs in the L and H lines, respectively.

In the first selected generation (G1) the Bs were transmitted on the female side (female $2B \times$ male 0B crosses); the L line showed strong B loss (mean number of Bs per plant in the progeny (mBs) = 0.55), whereas in the H line the Bs were strongly accumulated (mBs = 1.85). In the second selected generation the Bs were transmitted by the male (female $0B \times$ male 2B crosses); the L line transmitted the Bs nearly at a Mendelian rate (mBs = 1.03), whereas they were accumulated in the H line (mBs = 1.33).

The scheme of the crosses studied in the present work (G3) is shown in Fig. 1.

In plot 1, both the 2B and 0B plants of G3 came from a female 0BL × male 2BL cross of G2 showing 0.54 Bs per plant in the progeny; therefore, both the Bs and the As belong to the L line.

In plot 2, both the 2B and 0B plants of G3 came from a female 0BH × male 2BH cross of G2 showing 1.56 Bs per plant in the progeny; therefore, both the Bs and the As belong to the H line.

In plot 3, both the 2B and 0B plants of G3 came from a female 0BL × male 2BH cross of G2, showing 1.37 Bs per plant in the progeny; therefore, the Bs belong to the H line, whereas the As are LH hybrids.

In plot 4, both the 2B and 0B plants of G3 came from a female 0BH × male 2BL cross of G2, showing 0.82 Bs per plant in the progeny; therefore, the Bs belong to the L line, whereas the As are HL hybrids.

In every plot 12 plants were sown, six 0B to act as pollinators and six 2B were emasculated to be used as females, with the exception of plot two where only five 2B plants were available. The offspring were collected from the emasculated plants of these female 2B × male 0B crosses.

Seedlings were scored for B chromosome number in root tips following fixation in 1:3 acetic acid:ethanol and staining by the Feulgen method. The statistical analyses were carried out using the STATGRAPHICS 6.0 computer program.

Results

The purpose of the present work was to determine if the genes controlling rye B transmission rate in the L and H lines are located on the A or on the B chromosomes. If such genes were on the Bs, the progenies would show low B transmission rates in plots 1 and 4, and high B transmission rates in plots 2 and 3 because the status of the parental Bs is L or H, respectively. However, if the genes were on the As, the progenies would show the same type of transmission in plots 3 and 4 because both are HL hybrids, whereas they would be of L type in plot 1, and of H type in plot 2.

Table 1 shows the B transmission obtained in plots 1–4. These results are in agreement with the hypothesis that the genes controlling rye B transmis-
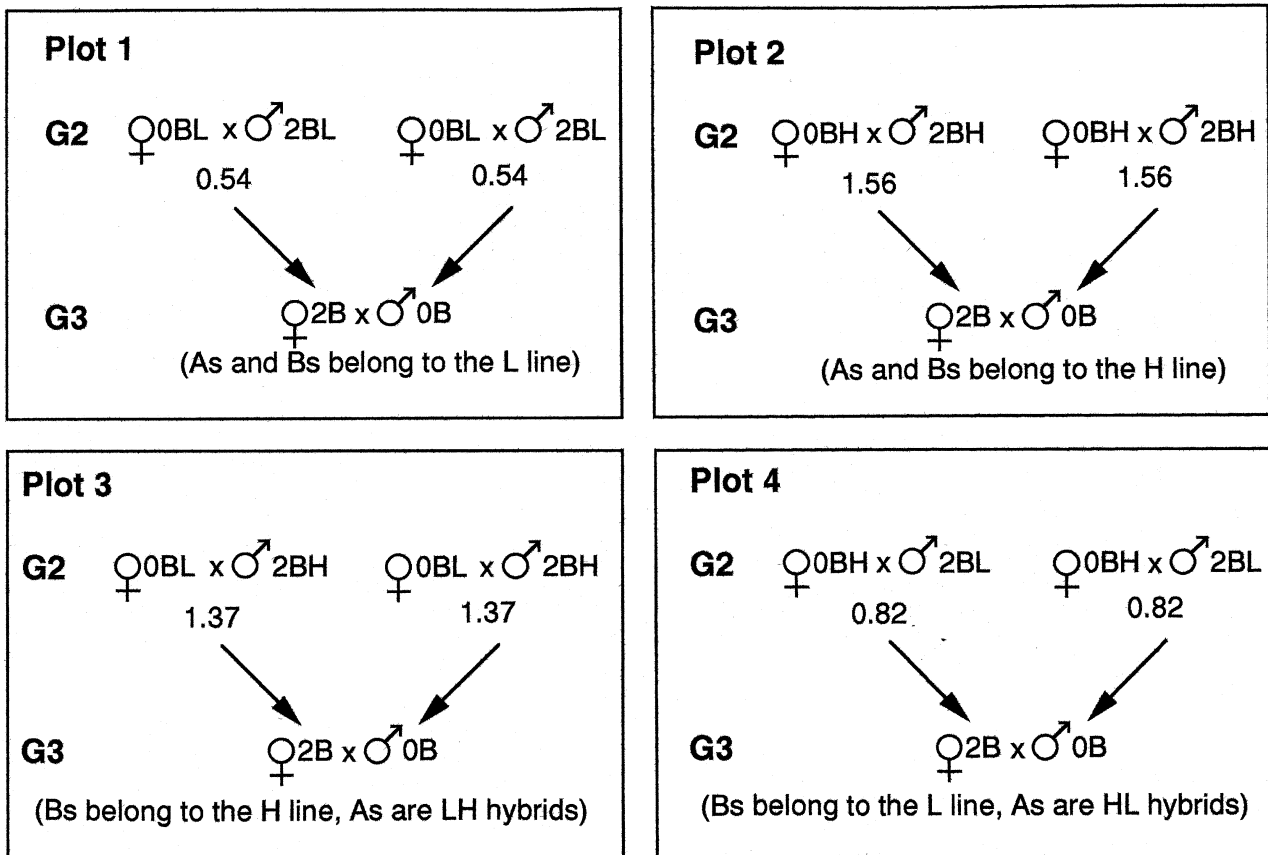

Fig. 1 Scheme of the *Secale cereale* crosses.

Table 1 Progeny obtained in female 2B × male 0B crosses involving rye plants of the high and low B transmission rate lines

Parents of the crosses in G2	No. of crosses in G3	B frequency in the progeny				Total no of plants	Mean no of Bs per plant
		0	1	2	4		
Female 0B L × male 2B L (plot 1)	6	85		53	4	142	0.86
Female 0B H × male 2B H (plot 2)	5	39	2	67	3	111	1.33
Female 0B L × male 2B H (plot 3)	6	53	3	94		150	1.27
Female 0B H × male 2B L (plot 4)	6	173	3	43		219	0.41

sion rate are located on the Bs because the progenies of plots 1 and 4 have a low mean Bs per plant (0.86 and 0.41, respectively) like their L 2B parents (Fig. 1), whereas the progenies of plots 2 and 3 show high mean frequencies of Bs per plant (1.33 and 1.27, respectively) like their H 2B parents (Fig. 1).

This conclusion is further supported by the following analysis. The mean number of Bs per plant in the 23 crosses summarized in Table 1 was compared by a two-way ANOVA (Table 2). The sources of variation were '0B female parent' and '2B male parent' because we compare the progeny of the 0B female parent, which transmits only the As in G2, and the male parent which transmits the As and the Bs in G2 (Fig. 1). If the genes controlling B transmission rate were on the As, we should find significant differences for both sources of variation, because the H or L genes of the As would be transmitted by both the 0B and 2B parent plants, whereas if the genes were on the Bs we should find significant differences only for the male parent, because this is the only source of H or L Bs in G2. The results of the ANOVA show that the effect of the 0B female parent is nonsignificant, whereas the effect of the 2B male parent is highly significant. The interaction between the sources of variation is also significant indicating that the source of the B chromosomes is not the only factor regulating their transmission rate.

Discussion

The results of this paper unambiguously show that the main difference between the high and low B transmission rate lines selected from the Puyo population is caused by 'genes' located on the Bs. The significant interaction between the main effects of the ANOVA (Table 2) suggests that the A chromosomes also affect transmission, but to a relatively small extent.

We have reported in previous papers that the low or high B transmission rate in crosses involving 2B and 0B plants is caused by the differential proportion of 0B vs. 2B gametes produced by the 2B plant and not by a different nondisjunction rate. On the other hand, the proportion of 0B and 2B pollen grains formed by a 2B plant depends mainly on B univalent or bivalent formation at metaphase I (Jiménez et al., 1995, 1997). According to these data, the simplest assumption is that the 'genes' for high or low transmission rate selected in the H and L lines are sites for chiasma formation located on the B chromosomes.

Table 2 Analysis of variance for the mean number of Bs in the progeny of plots 1–4

Source of variation	SS	d.f.	MS	F	P
Main effects					
A: 2B male parent	2.800	1	2.800	34.038	<0.0001
B: 0B female parent	0.272	1	0.272	3.311	0.0846
Interactions					
AB	0.432	1	0.432	5.253	0.0335
Residual	1.562	19	0.082		
Total (corrected)	5.093	22			

The B transmission rate did not increase in the H line, or decrease in the L line, from the first to the second and third generations of the selection process, indicating a high heritability and a low number of loci controlling the selected character, probably just one. It should be noted, however, that the selection method (2B × 0B crosses) does not permit the selection of the extremes of the distribution because the lowest end consists of all 0B progenies and the highest contains nearly all B carriers. We must select L progenies with some 2B plants because 0B × 0B crosses lack any information about B transmission. We selected H progenies with some 0B plants so that a similar selection process was used in both lines and because 2B × 2B crosses produce many 4B offspring which are difficult to maintain because of their low fertility.

Besides rye, there are a number of species where variation in B transmission rate has been reported: *Zea mays* (Carlson, 1969; Rosato *et al.*, 1996), *Hypochoeris maculata* (Parker *et al.*, 1982), *Myrmeleotettix maculatus* (Shaw & Hewitt, 1985; Shaw *et al.*, 1985), *Pseudococcus affinis* (Nur & Brett, 1985, 1987, 1988), *Crepis capillaris* (Parker *et al.*, 1989), *Locusta migratoria* (Viseras *et al.*, 1990) *Nasonia vitripennis* (Beukeboom & Werren, 1993) *Aegilops speltoides* (Cebriá *et al.*, 1994), *Allium schoenoprasum* (Bougourd & Plowman, 1996) and *Eyprepocnemis plorans* (Herrera *et al.*, 1996). The finding that the genes controlling rye B transmission rate are located on the Bs is unexpected because most previous reports, including our own (Puertas *et al.*, 1993), demonstrate or assume that transmission rate modifier genes are located on the As. But few studies unambiguously locate transmission rate modifier genes and the existence of genes located on the Bs is usually untested. For example, Nur & Brett (1987) compared males carrying the same B and the same paternal genome but differing in their maternal genome; they demonstrated the existence of transmission rate modifier genes on the As, but possible variation in the Bs was not tested.

The results of the present paper give us a new view of the genetic mechanisms which maintain rye Bs in populations. If the genes controlling B transmission rate are located on the Bs we could think of the Bs controlling and moderating their own presence in rye populations.

The mechanism of rye B accumulation is remarkably strong and rye is the only species where the 'drive' has been shown to occur on both the female and male sides. Moreover, in all studied rye populations, B-directed nondisjunction occurs in nearly 100 per cent of the pollen grains having the B (Romera

et al., 1989). This strong drive is thought to be counteracted by the harmful effect of a high number of Bs on the fitness of the carrier. In addition, it might also be counteracted by the tendency of the Bs of the L-type to be lost because L 2B plants produce many 0B gametes. L-type carriers would be favoured by natural selection because they would suffer less from the B's deleterious effects. However, this apparent benefit may be counteracted by another harmful effect, because L 2B plants produce a high percentage of inviable pollen grains compared to the H 2B ones (Jiménez *et al.*, 1977).

Considering that the L and H Bs may be at different frequencies in different populations and that the carriers may have different fitnesses, it would be possible to maintain stable polymorphisms when the drive/loss balance is reached. Conversely, it might be possible for one type of B to be replaced by the other. According to Camacho *et al.* (1996) new selfish variants can appear from time to time replacing the former B version because of their superior transmission performance. It is also possible that there are different explanations for every species, either because the Bs have different evolutionary behaviour in different species or because the extant Bs are at different stages of the evolutionary process in each species or population.

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