found in several species (see Battaglia, 1964 for references). The influence of environmental factors on chromatin elimination induced by B's remains to be demonstrated, as does the likelihood of somatic variation in numbers of B's between main stalk and tiller. Experiments have been set up to resolve these questions.

M. M. Rhoades
Ellen Dempsey

3. Effects of various segments of the B chromosome on recombination and nondisjunction.

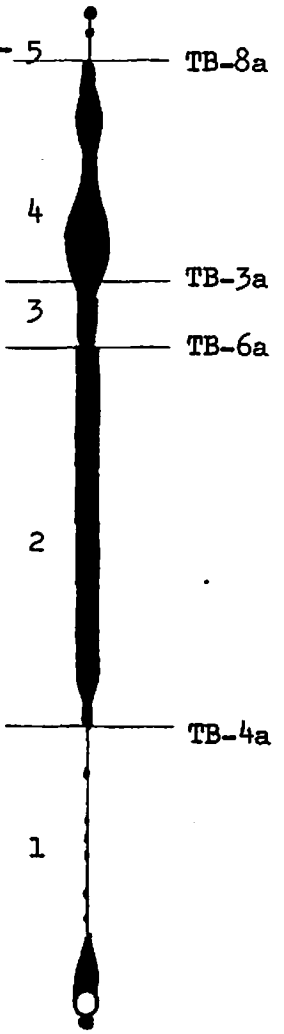
A) Analysis of the B chromosome segments responsible for enhanced crossing over.

Hanson studied recombination in chromosomes 9 and 3 and reported slightly higher values in plants with B chromosomes. These effects were not discernible until about four B's were present in the plant, at which time most of the enhancement was in double crossover classes. Since then Nel has found that B's appreciably augment exchanges in the centromere regions of chromosomes 5 and 9. The most spectacular promotion of recombination by B chromosomes, however, was discovered by Rhoades. A segment of 3L was transposed to the short arm of chromosome 9 and intercalated between C and Wx. Crossing over between the chromosome 9 markers was little affected when the transposition was homozygous, even though the physical distance was extended. This situation was drastically altered when a single B was present. Recombination was increased as much as 110% by the addition of a B chromosome, and a dosage effect was evident. The transposition line was utilized in crosses with selected A-B translocations in an attempt to determine which portion(s) of the B chromosome was involved in the enhancement of recombination.

Translocation stocks TB-4a, TB-3a, TB-6a, and TB-8a were made homozygous for the transposition and heterozygous for the markers C and Wx. Chromosomal constitutions of the resulting families were determined by pachytene analysis, root tip chromosome counts, and/or pollen abortion. The results of testcrosses are given in Table 1, and B chromosome breakpoints are shown in the accompanying drawing.

Table 1. Testcross data from C Tp Wx/ c Tp wx female parents varying in TB-A constitution, and a drawing of the B chromosome with breakpoints indicated by horizontal lines.

| Total population | % C-Wx Recomb. | Constitution |
|---|---|---|
| 2030 | 26.5 | $88^{B}$ |
| 1861 | 27.3 | 88 |
| 4597 | 32.4 | $88B^{8*}$ |
| 3499 | 38.6 | $88^{B}B^{8}B^{8**}$ |
| 1486 | 21.5 | 33 |
| 3059 | 30.2 | $33B^{3**}$ |
| 3115 | 36.0 | $33^{B}B^{3**}$ |
| 792 | 16.8 | 66 |
| 4048 | 27.7 | $66B^{6*}$ |
| 6550 | 31.3 | $66^{B}B^{6**}$ |
| 5704 | 37.6 | $66^{B}B^{6}B^{6**}$ |
| 3154 | 22.4 | 44 |
| 3169 | 28.7 | $44B^{4**}$ |
| 2395 | 33.9 | $44^{B}B^{4*}$ |
| 4414 | 38.2 | $44^{B}B^{4}B^{4*}$ |




*,** values are significantly greater than those in the compound immediately above at the 1% (*) and 0.1% (**) levels.

The lack of any increase in C-Wx recombination in the $88^{B}$ plants in relation to the control 88 class indicates that the distal-most region of the B chromosome, containing only two chromomeres, has no activity in the promotion of crossing over. As might be expected, the

$B^8$ chromosome is an effective inducer when only one is present, and two $B^8$'s enhance exchange even further. The higher recombination in the $33^B B^3$ class than in the $33B^3$ category indicates that the region of the B between the breakpoints of TB-3a and TB-8a is able to influence crossing over. The participation of the region of the B proximal to the TB-3a breakpoint is demonstrated by the promotion of crossing over by $B^4$, $B^3$, and $B^6$. The increase noted with the $B^4$ chromosome indicates that the euchromatic segment of the B (region 1) probably has activity in this phenomenon (the short arm or the centric heterochromatin of the long arm could be the center of activity). The heterochromatin between the TB-4a and TB-3a breakpoints (regions 2 + 3) may contribute to the enhancement of recombination or the $B^3$ and $B^6$ chromosomes could derive their activity from the presence of region 1 alone.

In an attempt to determine if segments 2 + 3 increase crossing over, the relative contributions of each portion of the B were estimated. Since the translocation lines were from different backgrounds, direct comparisons of recombination could not be made. However, if the exchange increment of the balanced translocation heterozygote above the control (with no translocation and no intact B's) represents 100% of the effect of an intact B chromosome, the percent contribution of each $B^A$ chromosome can be estimated by: $AAB^A - AA \times 100 / AA^B B^A - AA$, where AA, $AAB^A$, and $AA^B B^A$ are the percent recombination in plants of that constitution. The results of these calculations are as follows:

$$B^4 - 55\%;\ B^6 - 75\%;\ B^3 - 60\%;\ B^8 - 100\%.$$

The $B^4$ chromosome, containing only the proximal euchromatin of the B, has about 55% of the activity of an intact B chromosome. The 75% contribution of $B^6$ therefore suggests that 55% of the enhancement effect is due to region 1, while about 20% of the activity resides in region 2. The remaining contribution to the enhancement of crossing over by a $B^8$ or intact B must come from segments 3 + 4.

The 60% contribution to the enhancement of crossing over by $B^3$ is difficult to understand. With the breakpoint more distal than $B^6$, one might expect that $B^3$ would cause a greater increase in recombination than $B^6$. The region between the TB-6a and TB-3a breakpoints (region 3) may be inhibitory to crossing over and may counteract the stimulatory

effect of the more proximal regions. Whether or not an antagonistic condition exists, it is difficult to assign a percentage contribution to the segments studied. The most that can be stated at this time is that all segments of the $B_9$ with the exception of the distal chromomeres, appear to affect recombination in Tp9Tp9.

Since as yet no reliable data have been obtained on the effects of trisomics on recombination in the tested segment of chromosome 9, the possibility remains that some of the enhancement attributed to the $B^A$ chromosomes may be due to partial trisomy for the A chromosome involved in the translocation. Recombination in one trisomic 4 plant was not higher than that found in a closely related disomic. It is therefore likely that the $B^4$ enhancement of crossing over in Tp9Tp9 is a result of the presence of the B chromosome segment and not trisomy for chromosome 4 chromatin.

B) Control site for B chromosome nondisjunction.

With the establishment of the B chromosome breakpoint for TB-8a it became obvious that the opportunity existed to test the activity of the few distal chromomeres in the control of B chromosome nondisjunction. If the controlling segment lies in the region distal to the breakpoint, nondisjunction of the $B^8$ would occur only in $8^B B^8$ spores. However, if activity is confined to a portion proximal to the TB-8a breakpoint, the $B^8$ chromosome would undergo nondisjunction in both $8B^8$ and $8^B B^8$ spores.

Plants of constitution $88B^8$ were crossed as pollen parent by normal (88) individuals and chromosome counts were made on root tips of the progeny. The results are given in Table 2. In a total of 507 seedlings, 431 had 20 chromosomes, 73 had 21 chromosomes, and 3 were mosaic for chromosome number. Of the latter three, cells with 20, 21, and 22 chromosomes were found in two of the plants while the remaining seedling had cells with either 21 or 22 chromosomes. No plants with 22 chromosomes were found. The transmission of the $B^8$ through the pollen varied considerably, but averaged about 30%.

A further test for nondisjunction allowed for selection of seedlings which contained the $B^8$ chromosome, and made root tip analysis of large populations unnecessary. The procedure was as follows:

$8^v8^v \times 8^v8^v{}_B{}^{8V}$

$\downarrow$

$8^v8^v$

$8^v8^v{}_B{}^{8V}$ - from normal disjunction

$(8^v8^v{}_B{}^{8V}{}_B{}^{8V})$ - could arise from nondisjunction

Only green seedlings were analyzed for chromosome number, and they were expected to have 21 chromosomes if nondisjunction did not occur. Seedlings having two $B^8$ chromosomes would result from nondisjunction at the second microspore division followed by fertilization of the egg by the hyperploid sperm. Fourteen green seedlings were found in a total population of 119; all had 21 chromosomes.

When the data are pooled from both experiments, there were 87 plants having a single $B^8$ plus three seedlings in which somatic nondisjunction was occurring. Nondisjunction at the second microspore division was not observed in $88B^8$ plants. However, nondisjunction did occur with a high frequency in $8^B{}_B{}^8$ spores of $88^B{}_B{}^8$ and $88^B{}_B{}^8{}_B{}^8$ plants.

The most tenable conclusion from these experiments is that the control of B chromosome nondisjunction resides in the short distal euchromatic portion, containing only two chromomeres. It is interesting to note that this segment had no effect on recombination and thus these two phenomena are controlled independently.

Table 2. Root tip chromosome numbers from progeny of $88B^8$ males.

| Male plant | Progeny chromosome number | | | Total |
|---|---|---|---|---|
| | 20 | 21 | * | |
| 461-4 | 76 | 20 | 0 | 96 |
| 728-49 | 35 | 2 | 0 | 37 |
| 728-33 | 102 | 17 | 1 | 120 |
| 728-43 | 29 | 9 | 0 | 38 |
| 728-25 | 52 | 2 | 0 | 54 |
| 728-8 | 85 | 23 | 2 | 110 |
| 728-65 | 30 | 0 | 0 | 30 |
| 728-61 | 22 | 0 | 0 | 22 |
| Total | 431 | 73 | 3 | 507 |

*root tips were found to be mosaic for 20-21-22 chromosomes in two plants and 21-22 in the other.

C) A new observation of the effects of B chromosomes on recombination.

The recombination in the balanced heterozygotes of each of the TB-A translocations in section A can be considered to be equivalent to the crossing over occurring in Tp9Tp9 when a single intact B is present in the cell. The percentage increases from the AA class to the $AA^{B}_{B}{}^{A}$ group for each translocation were calculated and entered in Table 3. No balanced heterozygotes were successfully testcrossed in TB-8a, but absence of an effect on recombination by the distal chromomeres indicates that the $B^{8}$ has 100% of the activity of an intact B, and plants of $88B^{8}$ constitution were utilized in place of the balanced heterozygote. The data in Table 3 demonstrate that the lower the control value the greater the effect of the B chromosome. Regardless of the amount of crossing over in the controls, C-Wx recombination is usually increased to about 33-34%. Rhoades' results with OB vs. 1B classes are not inconsistent with these observations. Recombination in his OB plants was 17.7%, increasing to 37% with the addition of a B. This represents a 109% increase.

Table 3. Comparison of the increase in recombination in Tp9Tp9 caused by the B chromosome as represented by the balanced heterozygotes of TB-3a, TB-4a, and TB-6a and by the $B^{8}$ of TB-8a.

| Constitution | % C-Wx Recomb. | % increase |
|---|---|---|
| 88 | 27.3 | 18.7 |
| $88B^{8}$ | 32.4 | |
| 44 | 22.4 | 51.5 |
| $44^{B}_{B}{}^{4}$ | 33.9 | |
| 33 | 21.5 | 67.5 |
| $33^{B}_{B}{}^{3}$ | 36.0 | |
| 66 | 16.8 | 86.5 |
| $66^{B}_{B}{}^{6}$ | 31.3 | |

Kikudome found a comparable situation when he studied the effect of K10 on crossing over in a chromosome 9 bivalent heteromorphic for a knobless chromosome and a small, medium, or large knob. Crossing over increased from 26.9% to 31.5%, from 17.7% to 26.8%, and from 12.7% to 30.3%, respectively. These results represent increases of 17.1%, 51.5%, and 140%. It was suggested that there was an upper limit of about 30% recombination in the region investigated. In tests of Tp9Tp9 recombination there is a dosage effect of B chromosomes; therefore, the 33-34% commonly found with one B cannot be an upper limit.

Edward Ward

4. Detection of somatic redundancy for the $Adh_1$ gene of maize: a genetic strategy and preliminary data.

With few exceptions, the genetic information specifying the unique amino-acid sequence of an enzyme-subunit polypeptide is transmitted through meiosis as a single cistron. A diploid cell contains "one" paternal and "one" maternal allele for each gene, where the number "one" connotes one unit of genetically transmissible information (allele), as opposed to one unit of biosynthesis (here called "cistron"). Certain somatic cells might contain many redundant copies (cistrons) of one gene, copies which are transmitted through mitosis, without violating Mendelian laws. Even gametes may be redundant for any genetically single allele if one invokes the speculative "master-slave" relationship (Callan, 1960; 1967) among the redundant cistrons. Cytological and biochemical techniques presently available lack the resolution needed to detect small amounts of redundancy. This short note outlines a genetic strategy theoretically able to discover redundancies between zero and about twenty copies of a specific enzyme-specifying allele, and gives preliminary data for one of the alcohol dehydrogenase genes ($Adh_1$; ADH enzyme, EC 1.1.1.1.) in maize root primordial cells. This strategy may have general applications.

Experimental strategy:

There are two alleles for each gene in a diploid somatic cell. If each allele were represented by but one cistron, then the maximum number of different polypeptides specified by a particular gene is two