If abnormal 10 has the ability to reduce the rate of nondisjunction of a B chromosome, an explanation may be advanced for Longley's data on distribution of B chromosomes in different maize stocks. (J. Agric. Res. 56:177). Longley found that B chromosomes were seldom present in stocks with large numbers of heterochromatic knobs. If knobs in general, and not just abnormal 10, are capable of reducing the rate of nondisjunction among B chromosomes, they would tend to eliminate B chromosomes from the stock. Nondisjunction of B chromosomes together with preferential fertilization of the egg by the hyperploid sperm increases the B chromosome number in a population. In the absence of nondisjunction, it seems likely that a lack of selective advantage and irregularities in segregation at meiosis would result in the elimination of B's. The effect of other knobs on the rate of nondisjunction in TB-9b will be tested this summer. There is no evidence yet that knobs other than abnormal 10 can affect nondisjunction.

Wayne Carlson

3. Further study of the transmission of B^4 derived from the TB-4a standard stock.

In last year's News Letter (1966) preliminary data on the transmission of the supernumerary B^4 in normal genotypes were reported. A more intensive study of its genetic behavior is underway. Crosses were made using hyperploid plants $(4,4,B^4)$ homozygous for Su as the male parent on a su_1 tester in order to investigate the transmission of B^4 together with chromosome 4 through the male gametophyte. In addition, the ability of such hyperploid gametes to fertilize the egg, as well as the nondisjunction rate of B^4 in the absence of $4B$ at the 2nd microspore division, was studied.

The F_1, heterozygous Su/su and expected occasionally to be hyperploid for B^4, was backcrossed as female parent to the su_1 tester in order to recover the hyperploid plants. Three hundred thirty ears were scored for the ratio Su:su. The presence of B^4 would be indicated by an excess of ears showing a significant deviation from the expected 1:1 ratio (with a majority of Su kernels) over those deviating by chance. Also, 19 ears obtained by selfing were scored for the ratio 3:1. A total of 108,273 kernels were classified.

In Table 1 the P value corresponds to the x^2 deviation calculated on each of the ears examined. These results show a considerable excess of ears deviating from the expected ratios. The 6 ears deviating from the 1:1 ratio in the opposite direction (towards su) give an idea of the deviation by pure chance. In the backcrosses, among the significantly deviating ears, variations of ratios from 1.88:1 to 1.30:1 are presumably due to different rates of loss of the single B^4 in female meiosis or in the later stages of embryo sac development. These variations could be partially ascribed to the different backgrounds originally involved. The few data available from the ears obtained by selfing confirm those available from backcrosses. Here pollen transmission of B^4 occurs in addition to the main source of transmission through the female gametogenesis, but at a low rate since the B^4-hyperploid pollen is competitively selected

Table 1

P values corresponding to the X^2 values calculated	No. of ears scored for the following segregations:			Total	Average ratio $\underline{Su} : \underline{su}$ (weighted values)	Average excess % of $\underline{Su}$ kernels
	1 : 1	>1$\underline{Su}$: 1$\underline{su}$	<1$\underline{Su}$: 1$\underline{su}$			
Backcross:						
P<0.001 (***)		28	0	28	1.88 : 1	46.7
P<0.01 (**)		11	2	13	1.46 : 1	31.5
P<0.05 (*)		9	4	13	1.30 : 1	23.0
P<0.05 (n.s.)	276			276	1.00 : 1	0.0
Total	276	48	6	330		
Self:	3 : 1	>3$\underline{Su}$: 1$\underline{su}$	<3$\underline{Su}$: 1$\underline{su}$	Total		
P<0.001 (***)		2	0	2	5.68 : 1	42.3
P<0.01 (**)		1	0	1	6.20 : 1	51.6
P<0.05 (*)		0	0	0		
P<0.05 (n.s.)	16			16	3.00 : 1	0.0
Total	16	3	0	19		

against in the presence of normal pollen. Nondisjunction of B^4 in the 2nd microspore division, when hyperploid plants $(4, 4, B^4)$ were crossed as male parent to $\underline{su}_1$, would have resulted sometimes in tetrasomic plants heterozygous for $\underline{su}$ ($4\underline{Su}$, $4\underline{su}$, $B^4\underline{Su}$, $B^4\underline{Su}$). These should segregate $\underline{su}$ at a very low rate in backcrosses, since in this double condition B^4 should be lost only at a low frequency. None of the 330 ears examined showed such a ratio. This does not mean absence of nondisjunction(according to Roman, 1949), since such tetrasomic plants, obtained from selfing hyperploid $(4, 4, B^4)$ genotypes, have a very low vigor and could have been lost.

In most of the backcrossed ears the excess of $\underline{Su}$ kernels was expected to be due to the presence of B^4 carrying the dominant $\underline{Su}$. Root tips from $\underline{Su}$ kernels were squashed with the standard Feulgen technique in order to score for the presence of B^4. This was found at a frequency closely corresponding to the excess of $\underline{Su}$ kernels:

Sampled progeny from a back cross of $4\underline{Su}$, $4\underline{su}$, $B^4\underline{Su}$ plants

	20 chromosomes	21 chromosomes	Total
Plants from Su kernels	13	8	21

The supernumerary chromosome observed in metaphase plates was a short telocentric chromosome, as expected for the B^4.

After selfing hyperploid plants $(4, 4, B^4)$ the following three genotypes were expected:

1. = 20 chromosomes

2. = 20 chromosomes + B^4

3. = 20 chromosomes + B^4 + B^4

The relative ratio of these three genotypes should give a preliminary idea about the transmissibility of B^4.

Microsporocyte samples were taken from 88 plants of such a progeny: 67 plants had 20 chromosomes and 21 plants had, in addition, one B^4; no plant was found having two B^4's (see 1966 M.N.L.)

After crossing the hyperploid plants found to $\underline{su}_1$ as mentioned above, 19 F_1 plants were selfed. Those heterozygous $\underline{Su}/\underline{su}$ and having in addition $B^4\underline{Su}^1$ gave rise to ratios near a 6:1. A sample of $\underline{Su}$ kernels has been planted, and root tips were scored from each of them.

Sampled progeny from self pollination of $4\frac{Su}{}$, $4\frac{su}{}$, $B\,4\frac{Su}{}$ plants

	20 chromosomes	21 chromosomes	22 chromosomes	Total
Plants from Su kernels	67	47	8	121

Most attention was drawn to the few plants with two B^4's. Nevertheless, because of their very low vigor (presumably due to the unbalanced genotype) it was not possible to make either self or sib pollinations. However, there was good pollen shedding, and a testcross was made:

$$(4\tfrac{su}{},\ 4\tfrac{su}{},\ B\,4\tfrac{Su}{},\ B\,4\tfrac{Su}{})\ \male\ \times\ \underline{su_1}\ \text{tester}\ \female$$

Two ears obtained gave rise to these data:

Ear no.	Su kernels	su kernels	Total	% su
1	113	19	132	14.4
2	161	15	176	8.5

The few su kernels are due to loss of $B\,4\frac{Su}{}$ when present in double condition in the plant. Scoring root tips from such kernels for the presence of B^4 will establish the rate of nondisjunction at the 2nd microspore division.

Most of the plants with one B^4 were selfed or backcrossed to $\underline{su_1}$. Since the plants could be either homo- or heterozygous at the su locus, different ratios were expected:

genotypes:	Progeny after selfing
$4\frac{Su}{}$, $4\frac{Su}{}$, + $B\,4\frac{Su}{}$	true breeding Su
$4\frac{Su}{}$, $4\frac{su}{}$, + $B\,4\frac{Su}{}$	segregating >3 Su : 1 su
$4\frac{su}{}$, $4\frac{su}{}$, + $B\,4\frac{Su}{}$	showing a low % of Su

The type true breeding for Su was actually found, and its recovery did not need a special attention.

The heterozygote expected to segregate in its progeny more than 3 Su to 1 su was examined in the following terms:

Ear no.	Su kernels	su kernels	Total	Ratio
1	164	23	187	7.14:1
2	136	19	155	7.16:1
3	305	88	393	3.47:1
4	352	66	418	5.34:1
5	211	33	244	6.43:1
6	226	36	262	6.28:1
7	398	62	460	6.38:1
Total: 7	1792	327	2,119	6.00:1 (aver. ratio)

The genotype expected to show a low percentage of Su kernels among the progeny was examined in these terms:

| Ear no.	Su kernels	su kernels	Total	% Su	Ratio
1	130	183	313	41.6	0.71:1
2	157	221	378	41.6	0.71:1
3	153	201	354	43.2	0.76:1
4	112	215	327	34.3	0.50:1
5	115	238	353	32.6	0.49:1
6	90	157	247	36.5	0.57:1
7	115	202	317	36.4	0.56:1
8	107	269	376	28.5	0.40:1
Total: 8	979	1,686	2,665	36.8 (aver. %)	0.60:1 (aver. ratio)

Data on backcrosses are not yet available. Su kernels taken from ears of the class segregating on the average 0.60:1 should have one of these genotypes:

$$su\ su + B^{4Su}$$

$$su\ su + B^{4Su} + B^{4Su}$$

$$Su\ su\ (+ B^{4Su})$$

The third class with B^4 not necessarily present, should be rare and due to occasional crossing over involving the segment of the B^4 from the su$_1$ locus to the breakage point and the homologous region of the chromosome 4 as indicated below:
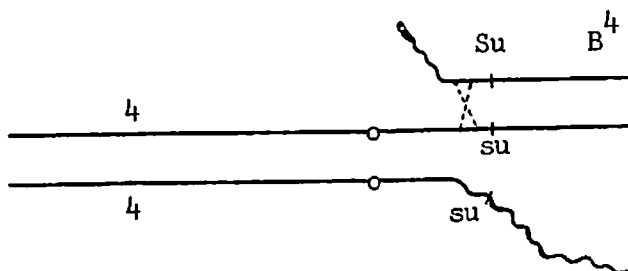
Fifty-six Su kernels were scored: 54 were found to have 21 chromosomes (presence of B^4) and 1 was found to have 20 chromosomes (no B^4). This could be the product of a rare crossover since in B^4 the breakage point is very close to su$_1$ (no more precise specifications have been reported to the author's knowledge); moreover, the pairing between specific segments of 4 and B^4, required for a crossover, occurs infrequently. However, the plant with 20 chromosomes could be a contaminant that was not detected since the stock is not provided with other markers. Another product of crossing over is a B^4su; this can only be detected cytologically among the larger class of su kernels. One plant in this sample was found to have 22 chromosomes (two B^4's); this indicates very low transmission of B^4 through the pollen.

Testcrosses were made of these genotypes:

$$(\underline{su}\ \underline{su}\ B^4\underline{\frac{Su}{}})\ ♀\ \ x\ \ \underline{su}_1\ \text{tester}\ ♂\ \ \text{and reciprocals}$$

in order to establish the transmissibility of B^4 through the meiotic barrier (the B^4 being single) and through the male gametophyte barrier (the B^4-hyperploid pollen grains being selectively disadvantaged when in competition with the normal ones). Data are not yet available, the material being in storage. Endeavors are being made to identify as many plants as possible with two B^4's in order to compare the kernel weight of classes having in the endosperm 0 B^4, one B^4, two B^4's, three B^4's and four B^4's. In the F_1 derived from pollinating inbred lines and hybrids with the TB-4a stock, the hyperploid endosperms were proven to be heavier (Bianchi, Genetics, 1962; Bianchi, Bellini and Ottaviano, Z. Vererb., 1962; Bellini, Ottaviano and Ghidoni, M.N.L., 1961).

Some conclusions can be drawn from the study of B^4 as a supernumerary in normal genotypes:

1. When B^4 is a single supernumerary it suffers severe losses during meiosis; and, when it escapes the meiotic barrier, such hyperploid

pollen is selected against because of competition with normal pollen (Ghidoni, Atti A.G.I., 1965; and M.N.L., 1966).

2. Hyperploidy for the B^4 results in poor growth of the plant which becomes more pronounced with increased doses.

3. When two homologues B^4 are present the transmission is still not ensured to all gametes; some meiotic losses are possible as well as a low rate of non-disjunction either during meiosis or in the microspore divisions. This seems to preclude the possibility of fixing lines with such specific hyperploidy, since this attempt of altering permanently the genotype results in a deleterious unbalance of genetic factors.

Achille Ghidoni

4. An improved method for detecting monoploids from different inbred lines.

A considerable number of methods have been used to screen for maternal monoploids. Most of them were suggested by S. S. Chase (PNAS, 1947 and Agr. J., 1952) and successfully carried out by E. H. Coe, Jr. (J. Her., 1964).

Recently A. Ghidoni and E. Ottaviano (Genetica Agraria, Proc. 1966) suggested (as did R. W. Briggs independently in the M.N.L. 1966) that the colored scutellum stocks, i.e., homozygous for $A_1 A_2 C R$ and the scutellum color markers (S_1, either two of the $S_2 S_3 S_4$ series, and the recessive s_5), can be used as male parent to pollinate several colorless inbred lines used as female parents. Monoploids can obviously be detected in those lines not carrying C^I, and carrying the recessive s_5. It should be easy to recognize in which lines these two conditions are satisfied, since:

1. the lines carrying C^I, pollinated as mentioned above, should have the F_1 fully colorless (endosperm and embryo);

2. the lines carrying S_5 (dominant) should have, in the absence of C^I, F_1 seeds with colorless scutellum only.

These two categories can be easily recognized and therefore discarded. Those showing both endosperm and scutellum color are to be scored for putative monoploids, which would have colored endosperm and colorless embryo. Crosses were made on 25 colorless inbred lines, but the colored scutellum stock used as the male parent was not homozygous for all factors and therefore no data were available. However, Briggs' data showed the effectiveness of the method.

This method of testing different lines as potential producers of monoploids is similar to those involving in the male parent either the dominant colored plumule factors ($Pu_1 Pu_2$) or the dominant "Purple Embryo Marker" (which is R^{nj} in a proper background) as they were developed by Chase and I. Greenblatt (M.N.L. 1965, 1966 and Crop Sci., 1966). However, it has the advantage that colored scutellum classification is normally less