

MAIZE GENETICS COOPERATION

NEWS LETTER

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I. FOREWORD

In our letter of 8 December, 1961, requesting material for the 1962 Maize Genetics Cooperation News Letter we stated that contributions should consist of notes and reports of research on genetic (broadly defined) studies. It was felt that the pages of the News Letter should be utilized primarily for reports of genetical interest and that we should not encourage the submission of contributions of a purely agronomic character, for otherwise the News Letter would become unmanageable in size. Although the decision as to the appropriateness of a report was left to the judgment of the individual investigator, it is our impression that this policy has resulted in an improved News Letter.

A considerable portion of the expense of publishing the 1962 News Letter was met by a grant from the National Science Foundation.

As has been true for the past six years the task of editing and assembling the News Letter has been in the capable hands of Miss Ellen Dempsey. We gratefully acknowledge our appreciation of her invaluable services.

M. M. Rhoades

II. REPORTS FROM COOPERATORS

BROOKHAVEN NATIONAL LABORATORY*
 Upton, New York
 Department of Biology

1. The effect of aneuploidy upon the chromosome number of succeeding generations in tetraploid maize.

Chromosome numbers were determined for individual plants in a population of tetraploid Argentine flint maize. Pollinations were made by bulked pollination in such a way that plants with 38 chromosomes were pollinated by bulked pollen from plants with 38 chromosomes, 39 chromosome plants by 39 chromosome pollen, etc.

The effect of parental chromosome number on the succeeding generation was determined by counting chromosomes in the root tips of seedlings resulting from the above procedure. The results are given in Table 1.

Table 1. Summary of chromosome numbers in the progenies of plants having several different chromosome numbers.

Parental chrom. no.	No. of parent plants	Progeny chromosome number								No. plts. ctd.	Av. no. chrom.
		37	38	39	40	41	42	43	44		
38	6	33.3%	16.7%	33.3%	16.7%	--	--	--	--	6	38.3
39	15	1.5%	20.6%	38.2%	35.3%	2.9%	1.5%	--	--	68	39.2
40	32	--	4.0%	10.7%	54.7%	21.3%	8.0%	1.3%	--	75	40.2
41	20	--	1.8%	7.3%	40.0%	23.3%	27.6%	--	--	55	40.7
42	15	--	--	4.9%	31.7%	24.4%	31.7%	4.9%	2.4%	41	41.1
43	3	--	2.5%	2.6%	10.5%	16.0%	34.2%	23.7%	10.5%	38	41.9

The t tests for skewness of the above distributions were as follows:

39 chrom. population t = +0.4931
 40 chrom. population t = +0.6993
 41 chrom. population t = -0.7651
 42 chrom. population t = +0.5780
 43 chrom. population t = -1.8410

None of these populations was significantly skewed, though the t value for the 43 chromosome population is near significance.

Next, all possible t tests were made to determine if average chromosome numbers of the progenies were significantly different from each other:

*Research carried out in part at Brookhaven National Laboratory under the auspices of the U.S. Atomic Energy Commission.

39 vs. 40	t = 6.024**
39 vs. 41	t = 7.980**
39 vs. 42	t = 8.887**
39 vs. 43	t = 11.250**
40 vs. 41	t = 2.839**
40 vs. 42	t = 4.599**
40 vs. 43	t = 7.692**
41 vs. 42	t = 1.817
41 vs. 43	t = 4.800**
42 vs. 43	t = 2.819**

** = significant at 1% level

Since all comparisons between mean chromosome numbers were significant, or nearly significant, it can be concluded that parental chromosome number has a pronounced effect upon that of the following generation, and this factor must be considered in evaluating the causes of sterility in autotetraploids.

Since even the progenies of parents with extreme chromosome numbers did not contain individuals beyond the range of 37 to 44, it is logical to conclude that embryos or gametophytes having numbers (or potentially having numbers) beyond this range are non-viable. It is a necessary corollary that aneuploidy per se has an important effect upon seed set in $4n$ maize.

Donald L. Shaver

2. The effect of structural heterozygosity on the degree of preferential segregation in allotetraploids of Zea ($4N$ Z. perennis x $4N$ Z. mays).

Polyploidy has undoubtedly played a major role in the speciation of the angiosperms, and has accounted for at least 30% of the extant forms. According to the most popular current theory, this course of evolution entails production of interracial (or wider) hybrids, doubling these to produce allotetraploids, and then (typically) the process of diploidization is completed by divergent genetic and structural modification of the newly combined genomes, until preferential pairing is essentially complete.

However, it does not appear that the actual effects of a defined structural modification on preferential segregation have been measured in an allotetraploid.

By means of genetically marked maize tetraploid stocks carrying Inversion 3a (obtained from Dr. G. G. Doyle), one can quantitate the effect of this structural rearrangement on segregation of the included linked markers a₁ - lg₂. These data are shown in Table 2.

Table 2. Gametic output of two allotetraploids of *Zea*, similarly marked, but differing structurally.

Allotetraploid	"Phenotypes" of gametes				No. of gametes
	A_1-Lg_2	A_1-lg_2	a_1-Lg_2	a_1-lg_2	
<u>a_1 Inv. 3a lg_2</u>					
<u>a_1 Inv. 3a lg_2</u>					
<u>A_1 Lg_2</u>	95.6%	0.6%	0.2%	3.6%	2003
<u>A_1 Lg_2</u>					
<u>a_1 lg_2</u>					
<u>a_1 lg_2</u>					
<u>A_1 Lg_2</u>	85.4%	1.7%	6.8%	6.1%	2146
<u>A_1 Lg_2</u>					

Seg. ratio for a_1 without inversion = 7.75:1
with inversion = 26.32:1

Seg. ratio for lg_2 without inversion = 12.82:1
with inversion = 23.81:1

The X^2 for the effect of the inversion upon the overall array of gametes is 464.356; $P = < .0005$.

For the effect of the inversion on segregation of a_1 , $X^2 = 147.370$, $P = < .0005$.

For the effect of the inversion on segregation of lg_2 , $X^2 = 36.038$, $P = < .0005$.

It can be concluded that the addition of structural divergence into newly formed allotetraploids would greatly increase preferential pairing and enhance the process of diploidization.

Donald L. Shaver

3. Trivalent frequencies in several interspecific hybrids of *Zea*.

Since preferential pairing in polysomics has been used as a measure of phylogenetic relationship, it is of interest to apply this measure among species of *Zea*. Triploid hybrids were made by crossing 4n *Zea perennis* (perennial teosinte) by 2n *Zea mexicana* (Florida teosinte) and 3 strains of 2n *Zea mays*. Since trivalent pairing in these hybrids is non-preferential, and univalent-plus-bivalent pairing is preferential, one can use trivalent frequency as a measure of the degree of preferential pairing. These frequencies are shown in Table 3.

Table 3. Trivalent frequencies in several interspecific triploid hybrids of Zea.

Triploid hybrid	No. plants studied	No. sets of homologues scored ¹	Trivalent Freq.
perennial teosinte x Fla. teosinte	1	1340	3.597 ± 1.344
perennial teosinte x Gaspé Flint	1	1380	6.594 ± 1.412
perennial teosinte x Cuzco Flour corn	1	1330	6.143 ± 1.634
perennial teosinte x Kys pure line	2	1400	6.857 ± 1.563

¹All data come from whole-cell analysis.

Taken at face value, these results lead to the surprising conclusion that the trisomic test for homology places maize and perennial teosinte closer together than perennial and annual teosinte. These data, however, need to be supported by observations from a larger number of plants. Moreover, it may be necessary to distinguish between the competition for pairing which is present in trisomics and the preference in pairing which is found in tetrasomics.

Donald L. Shaver

4. Perennial diploid Zea?

Perennial diploid Zea would be useful for many reasons. However, Randolph found that parthenogenetic diploids of $4n$ Zea perennis were rarely produced, and never viable. If this inviability is due to accumulation of protected lethals in the tetraploid, it should be possible to obtain diploids from this material by crossing the triploid hybrid of $4n$ Zea perennis x $2n$ maize back to $2n$ maize. Since autosyn-desis usually occurs in the $3n$ hybrid, it can be expected that diploids resulting from the backcross will be essentially F_1 's having one Z. perennis and one Z. mays genome.

Twelve diploids and near-diploids have been produced by this process. These are viable, produce flowers, and set seed. Their growth habit ranges from apparently perennial to annual.

It should be a straightforward matter to produce fully rhizomatous and perennial diploids by genetic recombination within this group. The proportion of maize chromatin could then be gradually increased by further cycles of crossing and recombination.

Donald L. Shaver

5. A classical test for allelism of id (indeterminant growth habit = photoperiodic) in teosinte and maize.

Three hybrids between maize which is homozygous id and teosinte were found to be indeterminant. They flower, however, in response to a photoinductive regime of 9 hrs. of light and 15 hrs. of dark.

This finding is a successful partial repetition of the work of Langham (Genetics 25:88-107, 1940), and is a partial reconfirmation of his contention that the inheritance of this difference between maize and teosinte is simple (monogenic).

Donald L. Shaver

BUTLER UNIVERSITY
Indianapolis, Indiana

1. Growth effects of gibberellic acid on dwarf-1 and normal maize seedlings.

One approach to the basic problem of the role of hormones in normal plant growth is to use artificial applications of hormones to plants in which the usual amount of native hormones is assumed to be reduced by mutant gene action. In maize a "growth series" can be set up using the mutant, d₁, and artificial applications of the hormone, gibberellic acid (GA₃), which has been demonstrated to normalize expression of this gene (Phinney, 1956, Nat. Acad. Sci. 42:185-189). The series is:

1. Normal phenotype (D₁D₁ and D₁d₁) untreated, assumed to be within the normal range of native hormone content.
2. Dwarf phenotype (d₁d₁) untreated, has shown evidence of reduced native hormones (Van Overbeek 1938 Plant Physiol. 13:587-598; Phinney 1961 Iowa State U. Press, pp. 489-501).
3. Normal treated with GA₃ -- normal native hormones and added GA₃.
4. Dwarf treated with GA₃ -- reduced native hormones plus added GA₃.

The maize kernels used in the present study were from the maize breeding program of Dr. E. C. Abbe of the Department of Botany, University of Minnesota. These kernels, segregating for d₁, were produced by back-crossing the mutant gene for several generations to University of Minnesota Station Inbred A188 to achieve a homogeneous background for the mutant gene. In the experiment daily applications of a 0.01% solution of GA₃ were applied to half the normals and dwarfs as soon as the dwarfs could be identified. Measurements of the first five leaves (which is about the life span of the treated plants) in maximum length and width of the leaf blade were made every other day until maturity of the leaves.

Three distinct patterns of growth could be detected from this growth series:

1. Normal growth as exemplified by the untreated normals.
2. Dwarf growth as produced by the untreated homozygous recessive mutants.
3. Extended growth resulting from additions of GA₃ to either the dwarf or normal phenotype.

Daily treatment with GA₃, then, changes both normal and dwarf growth. Early after treatment the dwarfs phenocopy the untreated normals, but soon the treated dwarfs copy the extended growth pattern such that the two phenotypes are indistinguishable. In terms of leaf form the three growth types are:

1. Normal growth characterized by long and narrow leaves.
2. Dwarf growth much shorter and wider leaves than type 1.
3. Extended growth much longer and narrower leaves than those of type 1.

Further experiments of this same nature will be made with larger populations and more frequent and complete measurements of leaf form than those used in this study.

Jeanette S. Pelton

THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION
New Haven 4, Connecticut

1. A sterile plant with S cytoplasm and S restorer genes.

In our 1961 MNL report (p. 20) on the genetic characterization of various sources of sterile cytoplasm it was pointed out that a single sterile plant appeared in the family from the cross A158H1♀ x (A158HxNY16)♂. By all tests source H is S type cytoplasm; NY16 contains S (and T) restorer genes. Thus, the heterozygous restored sterile A158H x NY16 would be expected to produce only fertile offspring when crossed as a male parent to the A158H sterile female, since in the presence of S cytoplasm pollen grains containing the nonrestoring allele abort and only the restorer allele is transmitted. It was suggested in the previous report that the exceptional sterile plant in the above test cross did have the major S restorer gene from NY16, but was not expressed in the residual genotype of the cross (which also contained 5 partially fertile plants). The single sterile plant, which was completely sterile and exerted no anthers, was pollinated by normal A158 (i.e. A158H1 (A158HxNY16) sterile plant x A158). In 1961 the progeny from this cross segregated 8 fertile and 9 sterile. Since A158 is free of S restorer genes, it is likely that the sterile female parent did in fact contain the S restorer genes.

Harry T. Stinson, Jr.

2. Segregation of T restorer genes in "reciprocal crosses."

In a previous MNL (1959; pp. 9-12) Jones reported that the heterozygous (Rf₁rf₁) restored sterile inbreds C103TF, HyTF and KrTF (TF = restored sterile) each produced a significant excess of fertile plants when crossed as pollen parents to T sterile single crosses. Genotypically all crosses were presumably T rf₁rf₁Rf₂Rf₂♀ x T Rf₁rf₁Rf₂Rf₂♂, and thus expected to segregate 1 fertile:1 sterile. The excess of fertile plants in these segregating progenies was attributed either to the presence of fertility modifier genes in the seed parent single crosses and in the pollinator inbreds, or to a selective mechanism favoring the Rf allele in the pollen. To obtain further information on the transmission and segregation of restorer genes, progenies from "reciprocal crosses" involving individual heterozygous restored sterile

plants were grown in 1960 and 1961. In making these reciprocal crosses a given restored sterile plant ($T \text{Rf}_1 \text{rf}_1 \text{Rf}_2 \text{Rf}_2$) was crossed as a male parent to a sterile female tester ($T \text{rf}_1 \text{rf}_1 \text{Rf}_2 \text{Rf}_2$), and as a female parent with the normal fertile version ($N \text{rf}_1 \text{rf}_1 \text{Rf}_2 \text{Rf}_2$) of the sterile tester (crosses symbolized as $T \times TF$ and $TF \times N$). As a check for the presence of genotypes which condition partial or full restoration in the absence of the major (Rf) restoring genes, progenies of the following types were also grown: sterile tester \times normal version of restored sterile; sterile version of restored sterile \times normal version of tester. Tassels were observed every other day, and classified as fertile (normal pollen production), sterile (no anther exerted), or partial fertile. The degree of fertility in the partial category was further estimated on the basis of the number of anthers exerted, the amount, if any, of pollen shed, and the time of anther exertion in relation to silking. The heterozygous restored sterile inbreds studied, the number of back-cross generations, and the source of the restorer genes (in parenthesis) were as follows: C103TF6(Ky21); HyTF6(C236); Oh51ATF6(IL53); A158TF5(Ky21). Results are summarized below for each of the four inbreds.

C103TF6 - Five different heterozygous plants in the BC6 were tested with three different sterile seed parents and their normal counterpart male parents (Table 1). None of the C103TF plants gave a significant deviation from the expected 1:1 segregation as either a female or male parent, nor did any of the reciprocal crosses differ significantly in the proportions of sterile and fertile plants. The combined results for crosses of the type $TF\text{?} \times N\text{?}$, as well as of the type $T\text{?} \times TF\text{?}$, give good fits to 1:1 ratios, and the five families in each group appear to be homogeneous. Finally, the reciprocal crosses, as groups, do not differ significantly, and their combined totals fit a 1:1 ratio. In all progenies (grown in 1960), segregation of fertile and sterile plants was clear, with no partially fertile plants present. The control crosses of the kind mentioned above contained only completely sterile plants.

HyTF6 - Five different restored sterile plants were tested with four different steriles and their normal counterparts (Table 2). HyTF plant No. 8, crossed with C103T12 and normal C103, gave a poor fit to a 1:1 ratio as both a seed and pollinator parent, and the two families were homogeneous in containing an excess of sterile plants. The control crosses, $\text{HyT} \times \text{C103}$, and $\text{C103T} \times \text{Hy}$, produced all sterile plants. The control crosses for the remaining four test crosses contained some plants classified as partial fertiles, which exerted a few anthers abnormally late and shed little or no pollen. Phenotypically similar partial fertile plants were observed in the segregating test crosses, and on the basis of the control crosses were classified as sterile. The remaining four HyTF plants gave relatively good 1:1 ratios, and in each case the segregations in reciprocal crosses did not differ significantly. However, the five families of the type $TF\text{?} \times N\text{?}$ give a combined segregation deficient in fertile plants, a deviation from expectation significant at the 5% probability level; the families satisfy the test for homogeneity. In contrast, the pooled segregations for the five families comprising the reciprocal cross ($T \times TF$) do not depart significantly from a 1:1 ratio, and these families also appear homogeneous. This difference in the behavior of the reciprocal crosses, as groups, borders on significance ($P = .06$). The results with HyTF are therefore inconclusive, but there may be a suggestion of a reciprocal cross difference. In any event, the results do not offer evidence for a significant excess of fertile plants when HyTF is used as pollinator.

Table 1 - C103TF6 crosses

Cross	F	S	P for 1:1	P for heterogeneity
C103TF6(15-6) x WF9	46	49	.75	
WF9T11 x 15-6	48	52	.70	
	<u>94</u>	<u>101</u>	.60	>.99
C103TF6(15-9) x (WF9 x 38-11)	61	48	.20	
(WF9T11 x 38-11) x 15-9	44	54	.30	
	<u>105</u>	<u>102</u>	.78	.12
C103TF6(15-14) x (WF9 x 38-11)	67	68	.87	
(WF9T11 x 38-11) x 15-14	60	57	.75	
	<u>127</u>	<u>125</u>	.85	.75
C103TF6(13-9) x (C106 x A158)	45	60	.15	
(C106T12 x A158) x 13-9	56	44	.27	
	<u>101</u>	<u>104</u>	.85	.07
C103TF6(14-5) x (C106 x A158)	62	66	.72	
(C106T12 x A158) x 14-5	59	69	.40	
	<u>121</u>	<u>135</u>	.38	.70
5 families combined:				
TF x N	281	291	.70	.40
T x TF	267	276	.70	.50
	<u>548</u>	<u>567</u>	.60	.78

Table 2 - HyTF6 Crosses

Cross	F	S	P for 1:1	P for heterogeneity
HyTF6(54-8) x C103	43	57	.17	
C103T12 x 54-8	33	47	.11	
	<u>76</u>	<u>104</u>	.04	.85
HyTF6(53-9) x (WF9 x W22)	41	49	.40	
(WF9T7 x W22) x 53-9	51	40	.35	
	<u>92</u>	<u>89</u>	.85	.20
HyTF6(54-4) x (WF9 x 38-11)	45	56	.28	
(WF9T11 x 38-11) x 54-4	91	82	.50	
	<u>136</u>	<u>138</u>	.87	.20
HyTF6(54-6) x (WF9 x 38-11)	39	45	.50	
(WF9T11 x 38-11) x 54-6	42	46	.70	
	<u>81</u>	<u>91</u>	.45	.85
HyTF6(53-1) x (C106 x A158)	40	48	.40	
(C106T12 x A158) x 53-1	57	51	.60	
	<u>97</u>	<u>99</u>	.70	.35
5 families combined:				
TF x N	208	255	.03	.98
T x TF	274	266	.70	.38
	<u>482</u>	<u>521</u>	.22	.06

A158TF5 - Three different restored sterile plants were crossed reciprocally with two different testers (Table 3). In five of the six families reasonably good 1:1 ratios were obtained; one family showed a poor fit. None of the reciprocal crosses differed significantly, and the pooled data for families of the type $TF^{\ominus} \times N^{\delta}$ and of the type $T^{\ominus} \times TF^{\delta}$ also do not deviate significantly from 1:1 ratios; the three families in each type of cross satisfy the test for homogeneity. The pooled reciprocal crosses also do not differ significantly from each other. There is thus no good evidence for abnormal segregations in the A158TF test crosses. The crosses with (WF9T x Oh51A) and its normal version contained a small proportion of partially fertile plants, but similar partial fertiles also appeared in the control crosses. These partial fertile plants were phenotypically similar to those reported above for the HyTF crosses, and were placed in the sterile category.

Oh51ATF6 - Only two restored sterile plants were tested, each with a different tester (Table 4). Each of the progenies involving one of the two Oh51ATF plants was deficient in fertile plants, and the reciprocal crosses were alike in this respect, each showing a significant departure from a 1:1 ratio. The second Oh51ATF plant produced a deficiency of fertile plants in one family and a significant excess of fertiles in the reciprocal cross, resulting in a highly significant difference in reciprocal crosses. The two crosses of the type $TF^{\ominus} \times N^{\delta}$ give a total segregation which deviates significantly from the expected ratio, each family being deficient in fertile plants. The two families of the reciprocal $T^{\ominus} \times TF^{\delta}$ cross are significantly different, one containing an excess of fertiles and the other an excess of steriles. The results with Oh51A restored steriles might be best explained on the assumptions that the single family containing an excess of fertile plants is exceptional, and that the two restored plants used in the crosses tended to produce a deficiency of fertile plants, perhaps because necessary modifier genes are lacking in certain offspring. However, none of the four families contained partially restored plants such as are frequently observed when modifier genes are segregating.

Summary: The above results indicate that the earlier finding of a significant excess of fertile plants in crosses of the type $T \underline{rf}_1 \underline{rf}_1^{\ominus} \times T \underline{Rf}_1 \underline{rf}_1^{\delta}$ is not a general phenomenon. Only one of the 15 families in this category contained a significant excess of fertile plants, and none of the pooled segregation ratios deviated significantly from expectation. Crosses with HyTF, however, did suggest a difference in reciprocal crosses, those of the type $TF^{\ominus} \times N^{\delta}$ showing a deficiency of fertile plants.

Table 3 - A158TF6 Crosses

Cross	F	S	P for 1:1	P for heterogeneity
A158TF5(69-1) x A	72	80	.50	
AT7 x 69-1	75	76	.90	
	<u>147</u>	<u>156</u>	.60	.70
A158TF6(69-4) x (WF9 x Oh51A)	94	70	.06	
(WF9T7 x Oh51A) x 69-4	63	53	.35	
	<u>157</u>	<u>123</u>	.04	.60
(A158TF6(69-5) x (WF9 x Oh51A)	85	90	.80	
(WF9T7 x Oh51A) x 69-5	59	47	.25	
	<u>144</u>	<u>137</u>	.70	.25
3 families combined:				
TF x N	251	240	.60	.15
T x TF	197	176	.28	.60
	<u>448</u>	<u>416</u>	.28	.60

Table 4 - Oh51ATF6 Crosses

Cross	F	S	P for 1:1	P for heterogeneity
Oh51ATF6(81-11) x (WF9 x Oh51A)	55	83	.02	
(WF9T7 x Oh51A) x 81-11	64	94	.02	
	<u>119</u>	<u>177</u>	<.001	.85
Oh51ATF6(81-3) x A	78	91	.30	
AT7 x 81-3	103	63	<.01	
	<u>181</u>	<u>154</u>	.14	<.01
2 families combined:				
TF x N	133	174	.02	.25
T x TF	167	154	.60	<.01

Harry T. Stinson, Jr.

3. Pollen transmission of T restorer genes by plants with sterile (T) and normal (N) cytoplasm.

A second aspect of restorer gene behavior investigated was a comparison of the segregation ratios produced by heterozygous (Rf_1rf_1) restorer male parents possessing T and normal (N) cytoplasm. The T and N restorer male parents were produced as follows: restored sterile lines of A158, Oh51A, and Kr which had been backcrossed 4 or 5 generations, selfed 3 or 4 generations, and shown by test crosses to be homozygous for the restorer genes ($T Rf_1Rf_1Rf_2Rf_2$) were crossed reciprocally with the respective normal lines (i.e. $N rf_1rf_1Rf_2Rf_2$) to give the two kinds of families, $T Rf_1rf_1Rf_2Rf_2$ and $N Rf_1rf_1Rf_2Rf_2$. In any given reciprocal cross the same two plants were used as parents. The original source of the restorer genes for A158 and Kr was Ky21, and the source for Oh51A was I153.

Pollen from the $T \underline{Rf}_1 \underline{rf}_1$ and $N \underline{Rf}_1 \underline{rf}_1$ (both are homozygous $\underline{Rf}_2 \underline{Rf}_2$) plants was applied to the silks of various T sterile testers. The sterile testers were also crossed with the normal ($N \underline{rf}_1 \underline{rf}_1$) lines of A158, Oh51A and Kr to determine the degree of restoration brought about by the genotypes of the test crosses in the absence of the \underline{Rf}_1 restorer gene. Progenies of the latter crosses contained either all sterile plants or sterile and fertile plants which produced little or no pollen. Consequently partial fertile offspring in the test crosses were classified as sterile.

Results are shown in Table 5. In the case of A158 there is no evidence for a difference in the behavior of A158NF and A158TF restorers, either in individual test crosses or in the combined results for the two types of crosses.

With Oh51A the crosses to C106T12 x A158 suggest a possible difference between the NF and TF restorers, the segregations in the families being almost exactly reversed. The combined totals for the T x NF and T x TF crosses do not give a significant X^2 for heterogeneity between the two kinds of crosses, but the families in the T x NF category give a poor fit for homogeneity.

None of the individual test crosses with the Kr restorers shows a significant difference between the NF and TF plants. The two kinds of crosses as groups also do not show a significant difference. However, the pooled T x TF crosses may contain a significant excess of fertile plants ($P = .03$), but the six families are of doubtful homogeneity ($P = .07$). Combining the results from all T x NF and all T x TF crosses gives 776 fertile and 700 sterile plants, a segregation with a P value of .05. There is thus a suggestion that Kr $\underline{Rf}_1 \underline{rf}_1$ restored steriles produce an excess of fertile plants when used as pollen parents, but there is no evidence for a difference between NF and TF restorers.

Table 5. A158N $\underline{Rf}_1 \underline{rf}_1$ and A158T $\underline{Rf}_1 \underline{rf}_1$

Sterile tester ♀	T x NF		P for 1:1	T x TF		P for 1:1	P for heterogeneity TxNF, TxTF
	F	S		F	S		
AT7	81	81	>.99	75	76	.93	.95
WF9T7 x Oh51A	70	59	.35	122	100	.14	.85
WF9T7 x W22	53	56	.90	88	80	.50	.60
Totals:	204	196	.70	285	256	.25	.60

P, heterogeneity,
families

.60

.55

Oh51A N Rf₁rf₁ and Oh51A T Rf₁rf₁

Sterile tester ♀	T x NF		P for 1:1	T x TF		P for 1:1	P for heterogeneity T x NF, TxTF
	F	S		F	S		
B8T7 x Oh43	85	74	.40	84	78	.70	.75
"	71	92	.10	77	81	.75	.35
WF9T7 x W22	85	83	.85	84	81	.80	.98
C106T12 x A158	71	92	.10	92	76	.22	.04
Totals:	312	341	.25	337	316	.40	.22

P, heterogeneity,
families

.15

.70

Kr N Rf₁rf₁ and Kr T Rf₁rf₁

Sterile tester ♀	T x NF		P for 1:1	T x TF		P for 1:1	P for heterogeneity T x NF, TxTF
	F	S		F	S		
WF9T7 x W22	74	60	.25	94	65	.03	.50
WF9T11 x 38-11	83	70	.30	80	77	.75	.60
C103T11 x Hy	76	80	.75	87	61	.03	.08
C106T12 x A158	74	74	>.99	78	81	.80	.85
C103T12	31	36	.60	32	42	.25	.85
KrT9	29	31	.85	38	23	.06	.12
Totals:	367	351	.55	409	349	.03	.27

P, heterogeneity,
families

.60

.07

Harry T. Stinson, Jr.

DEFIANCE COLLEGE
Defiance, Ohio1. Multiple conversion of R-locus expression in one generation.

R. A. Brink and his students have demonstrated that the expression of the R-locus can be altered by passing the R allele through a heterozygote with certain pattern alleles such as Rst and R^{mb} (stipple and marble). More recently, it was shown (PNAS 47:566) that R-locus expressions could be progressively converted to lighter and lighter phenotypes by passing R-alleles through pattern allele heterozygotes successively. That is, when RR^{mb} heterozygotes were crossed to RstRst(light) to yield in the next generation R'Rst and R'Rst(light) heterozygotes (prime is used here to indicate the number of pattern alleles with which R has been heterozygous), the testcrosses of these last heterozygotes gave R'' phenotypes which were significantly lighter than R' controls removed from the RR^{mb}, RRst and RRst(light) heterozygotes.

A further question remained; could the amount of change produced in two generations by the above method of progressive conversion be registered on the R-locus expression in a single generation by using more than one pattern allele.

A trisomic, $r^r R^{st} r^g_I$ (the allele r^g_I was a colorless mutant of Rst recovered by R. B. Ashman and capable of R-locus conversion), was crossed to the inbred RR homozygote to yield the $R^{st} r^g_I R$ trisomic as well as $R^{st} R$, $r^g_I R$ and $r^g_I r^g_I$ disomic heterozygote controls. The above trisomics and disomics were testcrossed for comparison of R' and R'' phenotypes. The amount of pigment produced in the endosperm was scored by matching kernels against a series of 23 standard kernels ranging from "zero" or colorless to complete pigmentation. Mean scores for 50 kernels from each ear are recorded below. The results show significantly lighter R' expressions from the trisomic as compared with any of the R' kernels from the disomic controls.

$R^{st} R r^g_I$	$R^{st} R$	$R r^g_I$	$R r^r$
<u>R</u> ''	<u>R</u> '	<u>R</u> '	<u>R</u>
3.26	8.24	6.52	18.96
4.10	8.98	11.28	18.60
4.64	6.76	10.10	19.80
3.66	10.86	16.04	19.00
2.06	9.58	8.52	18.00
3.08	11.12	10.36	20.12
4.80	10.86	11.86	
3.86	10.54	10.10	
3.56	13.98		
3.48			

Thus it is possible to influence R-expressions by progressive treatments over several generations or by using more than one pattern allele in a single generation.

Bernard C. Mikula and Steven D. Skopik

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1. Somatic crossing over ?

Cross:	$\frac{Su \ ga}{Su \ ga}$	x	$\frac{su \ Ga}{su \ Ga}$
F ₁ zygotes (expected):	$\frac{Su \ ga}{su \ Ga}$		

In one of three cultures containing F₁ plants of this origin were three plants which behaved in an unexpected manner. From selfing and from back-crossing (pollen to Ga ga su su plants) the following distributions were obtained:

Plant	Su	su	o/o su	P*	P* (comb. data)	Crossover** value
# 3 self	321	76	19.1	<.01		38.3%
BC	248	168	40.4	<.01	<.01	40.4%
#11 self	350	97	21.7	>.05		43.4%
BC	222	173	43.8	<.05	<.01	43.8%
#12 self	334	76	18.5	<.01		37.1%
BC	221	176	44.3	<.05	<.01	44.3%

* For deviations from 3:1 and 1:1

** Computed for coupling

Crossover values are rather high but comparable to those for high-sugary plants in the same culture.

Since all, or nearly all, of each tassel presumably was composed of cell descendants of a somatic crossover cell, it is suggested that the putative crossing over occurred early in ontogeny--possibly in zygotic mitosis.

Perhaps the use of a third gene such as Ts_p will clarify the situation by making possible a regular four-class backcross test.

H. S. Perry

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1. Further data on the components of the tunicate locus.

In a previous News Letter (No. 35) we reported that the two components of the Tu locus which had been separated by crossing over appeared to have slightly different effects. After additional backcrosses to the inbred A158 to render them more nearly isogenic, this is still true. In the summer of 1961 we grew seven strains heterozygous for tu^{h-d} , two of which were 13/16 A158 and five were 29/32 A158. These were compared with ten strains heterozygous for tu^{h-1} , three of which were 5/8 A158 and seven were 29/32 A158.

The two types of heterozygous half-tunicate genotypes could not be distinguished by their tassels or by the external characteristics of the mature ears, but examinations of longitudinal sections of both young and mature ears revealed differences. Pistillate spikelets from plants heterozygous for tu^{h-1} consistently had longer rachillae, slightly thinner lower glumes, and longer and more numerous cupule hairs than those from plants heterozygous for tu^{h-d} . In 71 paired comparisons of longitudinal sections of young ears the two types were consistently distinguishable. However, we cannot yet be certain that the differences are inherent in the two components and are not due to other genes closely linked with them.

P. C. Mangelsdorf

2. Combining chromosomes for Tripsacoid effects.

In an earlier News Letter (No. 32) I reported the extraction, from varieties of maize from the countries of Latin America, of chromosomes with effects similar to those of teosinte chromosomes. These were incorporated into the inbred, A158, through repeated backcrossing.

We have now intercrossed a number of the modified strains of A158 and grown F_2 progenies to determine whether the introduced chromosomes from different varieties are alike or different in their effects. If essentially the same chromosomes are involved in a cross there should be little segregation in the F_2 . If the chromosomes are different or carry different assemblages of genes, the F_2 should segregate and the F_2 population should include at one extreme ears quite similar to A158 lacking the introduced chromosomes and at the other extreme ears more Tripsacoid than either parent carrying both introduced chromosomes.

Of 31 F_2 populations involving extracted chromosomes from varieties from Mexico, Guatemala, Honduras, Nicaragua, Cuba, Venezuela, Brazil, Paraguay, Argentina, and Bolivia only two did not segregate. One of these was a cross of Argentina by Argentina which served as a control and the other a cross of Mexico by Honduras.

In a number of the F_2 populations the most Tripsacoid plants were barren, producing no ears or small cobs without grains, others had poorly formed ears which any practical corn breeder would immediately discard. Yet the F_1 plants from which these populations were derived were quite vigorous. Apparently this introduced germplasm, much of it probably originally from *Tripsacum*, is more or less beneficial when heterozygous but deleterious when homozygous. There is obviously a limit to the amount of foreign germplasm which can be introduced in a homozygous state into an inbred strain such as A158.

P. C. Mangelsdorf

3. Can proximity produce a heterosis-like effect?

Following a suggestion by J. B. S. Haldane, an experiment has been conducted to determine whether corn inbreds which produce marked heterosis when crossed together, can stimulate increased yield in one another when they are merely grown in close proximity. Haldane thought that such a reciprocal stimulation for higher yields between rice varieties grown together in a common flooded plot, as reported by S. K. Roy (1960), might have some relationship to heterosis.

Three types of plantings were made in four replications all involving two plant hills: (1) P39 alone; (2) A158 alone; (3) P39 and A158 together in pairs. The plantings were separated by adequate guard plots. The total yields of the two inbreds in the pure and mixed stands are shown below.

	Yield in grams			
	Pure stand	Mixed stand	Difference	t
P39	4245	6057	+1811	6.53* gain
A158	4460	3592	- 868	7.40* loss
Total	8705	9649	943	

*Both experimental t values are highly significant, being much larger than the tabular t at .01 of 2.71 (Snedecor Statistical Methods).

The results show that when P39 was paired with A158, the P39 component was more productive than when grown alone, but A158 with P39 was significantly less productive. Apparently P39 which has a tendency to tiller can compete more successfully than A158 which is single stalked. The net gain of 11 percent of the mixed stand over the pure stand is significant and may suggest an effect similar to heterosis, but the results are far from conclusive. A better experiment could probably be made by using two inbreds with similar or identical growth habits.

W. C. Galinat
P. C. Mangelsdorf

4. Effect of natural selection on teosinte introgression.

Various teosinte derivatives of A158 were intercrossed and a blend of the resulting seed was grown in isolation for four generations (years). Reserve seed from each year was planted in a 4 X 4 latin square yield test with the following results:

Generation	Yield bu/acre	Shelling %
1 (1957)	67.2	80.1
2 (1958)	70.8	77.1
3 (1959)	69.8	78.5
4 (1960)	66.6	78.1
For	0.05	8.7
Significance	0.01	13.1
		0.8

If the introgression of teosinte germplasm into corn causes evolution for increased yield, four generations of natural selection were inadequate to show it in the corn under the conditions involved in this experiment. The trial did show a significant drop in shelling percentage between the first and second generation.

W. C. Galinat
P. C. Mangelsdorf

5. Colchicine induction of an amphidiploid of multiple tester corn X *Tripsacum dactyloides*.

Among the many techniques and dosages for colchicine induction of polyploidy which were tried, only one was successful in producing the desired amphidiploid of a WMT corn X *T. dactyloides* hybrid. The successful procedure was as follows: A tiller about 18 inches long with adventitious roots starting to develop near its base was cut and grown in a nutrient solution until well rooted. The plant was then transferred to a mixed solution of aqueous colchicine (1:1000) and a non-ionic wetting agent (Tergitol 1:500) for 72 hours. The plant which appeared to be almost dead after this severe colchicine treatment, was transferred to a soil-Sphagnum mixture. After two months of being nursed along, a fairly normal cluster of seven shoots had emerged.

Stomata measurements suggested that three of these seven shoots were amphidiploid and this was later confirmed in chromosome counts made by Mr. Raju. This amphidiploid is fully female fertile on backcrossing to corn.

W. C. Galinat

6. High female fertility in F_1 hybrids of corn X *Tripsacum floridanum* and their backcrosses to corn.

Not only is *T. floridanum* highly crossable with some strains of corn, as I reported in last year's News Letter, but we now know that the F_1 hybrid and its backcross to corn are highly female fertile--seed set in the F_1 was 85% and almost this high in the backcross to corn. This discovery represents an important break through in both theoretical and applied work on the past and potential evolution of corn.

Such high female fertility in the F_1 and backcrosses to corn would make it easy, once a cross had occurred, for *Tripsacum* introgression into corn to occur in the wild or under conditions of primitive agriculture. It also makes the natural derivation of teosinte from such introgression seem more credible than some suggest. In this connection, we have already hybridized and backcrossed this most primitive species of *Tripsacum* with one of the most primitive living races of corn,

Confite Morocho, in an attempt to synthesize teosinte through controlled crossings. Also we are studying the inheritance of recessive marker genes of corn in corn-*Tripsacum* hybrids; this should lead to the development of a genetic map of *Tripsacum*.

A quantity of OP seed from an F_1 hybrid of A158 gl_3 X *T. floridanum* is available to those who wish to make use of it. This new source of germplasm should be especially valuable to those who are looking for new genes not presently available in corn.

Seed of *T. floridanum*, *T. dactyloides* 2n of Kansas and *T. dactyloides* 4n of Florida is also available.

W. C. Galinat

7. Chromosomes of three Mexican teosintes.

As previously reported, by crossing Mexican teosintes to a standard inbred strain of Wilbur's flint with virtually knobless chromosomes, the characteristics of the teosinte chromosomes can be determined by studies of the microsporocytes of the F_1 hybrid plants. During the past year, the following observations have been made.

Arcelia teosinte. Seed of this teosinte was collected near Arcelia, Guerrero. Of 21 F_1 hybrids of Wilbur's flint and Arcelia teosinte, only a few of the plants had good spreading pachytene chromosomes. As long as the bivalent pachytene chromosomes were clear and isolated, they appeared in close and regular association. With respect to knobs, there were two types of chromosome 1, one having a small internal knob on the short arm, the other having in addition two small internal knobs on the long arm. Chromosome 2 had two medium-sized internal knobs, one on each arm. Two types of chromosome 3 were observed, one knobless, the other with a large internal knob on the long arm. Chromosome 4 also had two types, one with a large knob on the long arm, the other, this knob and a small terminal knob on the short

arm. There was only one type of chromosome 5, having a small internal knob on the long arm. Two types of chromosome 6 occurred both with a short arm terminated by a small knob, but one having an additional medium-sized internal knob on the long arm. Chromosome 7 had a large internal knob on the long arm. There were two types of chromosome 8, one having two medium-sized knobs on the long arm, the other only one. Chromosome 9 had a medium-sized knob terminating the short arm. Two types of chromosome 10 were identified: one knobless, the other with a medium-sized internal knob on the long arm. The average length of the knobbed chromosome 10 is about four micra longer than that of the knobless one.

No inversions or any other gross chromosome rearrangements were observed in *Arcelia teosinte*.

Chilpancingo teosinte. Seed of this teosinte was obtained near Chilpancingo, Guerrero. Microsporocytes of 25 F_1 hybrid plants of Chilpancingo teosinte and Wilbur's flint were examined. At pachytene, bivalent chromosomes were closely associated. No chromosome rearrangements of any kind were found. The knob positions of this teosinte were as follows: There were three types of chromosome 1; that most frequently observed was knobless, the second most frequently observed type had a medium-sized internal knob on the long arm, and the type least frequently observed had three medium-sized internal knobs, one on the short arm, two on the long arm. Chromosome 2 had also three types: The first type had a medium-sized internal knob on the short arm, the second, an internal knob of the same size on the long arm, and the third had two medium-sized internal knobs, one on each arm. There were two types of chromosome 3; one had a medium-sized internal knob on the long arm, the other a large knob terminating the short arm. Chromosome 4 had a medium-sized knob terminating the short arm. There were two knobs on both types of chromosome 5; one had these knobs on the long arm, the other, on the short arm. These knob positions on chromosome 5 are new for teosinte. Chromosome 6 had three small knobs, a terminal one on the short arm and two internal ones on the long arm. There were two types of chromosome 7, one knobless, the other, with a large internal knob on the long arm. Chromosome 8 had two types; one had a medium-sized internal knob on the long arm, the other, a small terminal knob on the short arm. There were also two types of chromosome 9; one had a large terminal knob on the short arm, the other had in addition a small internal knob on the long arm. Chromosome 10 was knobless.

Perennial teosinte. Seed of perennial teosinte came from a stock obtained originally near Guadalajara, Jalisco. Microsporocytes of seven F_1 hybrid plants of perennial teosinte and Wilbur's flint were studied. All were found to have 30 chromosomes. No diploid or tetraploid plants were identified although tetraploid F_1 's had been previously obtained in the same cross by several earlier workers. At pachytene chromosomes were extremely entangled, as has been observed in the other triploid maize-teosinte hybrids. Chromosome pairing was irregular. A medium-sized internal knob on the long arm of chromosome 4 and a large terminal knob on the short arm of chromosome 7 were identified in two of the hybrids. In addition, practically all of the chromosomes had large chromomeres terminating one or two arms. A loop configuration of In9 was once clearly seen at pachytene. The length of the inverted segment is equivalent to about 60 per cent of the length of the short arm of chromosome 9, but the inversion figures were not abundant at

pachytene. This is perhaps due to the fact that teosinte chromosomes pair preferentially with teosinte chromosomes. Since teosinte contributes two homologues to each group of three chromosomes in the triploid, this In 9 would frequently occur as a homozygote, in which loops or other figures at pachytene, would not be expected.

At diakinesis, chromosome associations in a total of 101 randomly selected sporocytes were studied. The average numbers of trivalents, bivalents and univalents per sporocyte were 6 III, 4 II, 3.6 I. This type of association was found in 24 sporocytes in a total of 101. Twice in this total all 30 chromosomes associated into 10 regular bivalents. The manner of association of the three homologues in a trivalent ranged from: (1) end-to-end, (2) one chromosome attached to a parallel-paired bivalent, to (3) all three homologues paired in parallel fashion. The occurrence of bivalents was very common and probably most of them are formed by autosynopsis.

Despite the identification of In 9 at pachytene, bridges and fragments were not found at either anaphase I or anaphase II in the limited number of sporocytes examined.

Y. C. Ting

8. Evidence of the interchromosome effect of inversions on crossing over.

Data on the interchromosome effect of inversions on crossing over in plants are lacking, although data on this effect in Drosophila are abundant and show that when inversions exist as heterozygotes, they increase the frequency of crossing over in nonhomologous chromosome pairs (cf. Schultz and Redfield, 1930, 1951, Steinberg, 1936, Steinberg and Fraser, 1944, and Carson, 1953.) The affected nonhomologous chromosome pairs are either with or without any inverted chromosome segment. Last year, in microsporocytes of an F_1 plant (60-1105-1) of Kochimilco teosinte X Wilbur's flint, it was found that In 3 was present in addition to In 8 and In 9. This In 3, on the long arm of chromosome 3, was found to have an interchromosomal effect on both In 8 and In 9. Sporocytes of this hybrid had a high percentage of inversion configurations, practically all loops, of both In 8 and In 9 at pachytene. Among 215 randomly chosen sporocytes, 83, or 39 percent, had loop-configurations of either In 8 or In 9, or both. This frequency is much higher than that in plants which had In 8 and In 9 but not In 3, and which usually had not more than 10 percent of the sporocytes showing loop-configurations. The increase in the frequency of inversion loops, may be a good indication that the crossing-over frequency is likewise increased, since crossing-over within the inverted segments can happen only when the homologues are associated in loop-formations.

The second evidence of this effect was observed at anaphases of the sporocyte division. As shown in Table 1, about 13 percent of the sporocytes possessing In 3, In 8 and In 9, show evidence of crossing over at anaphase I.

In contrast to this among more than 500 sporocytes counted at anaphase I, a sporocyte without In 3 and having a dicentric bridge and an acentric fragment was found only once. At anaphase II about 3 percent of the daughter cells carrying In 3, In 8, and In 9 show evidence of

crossing over (Table 1). This is much higher than the percentage found in the sporocytes having In 8 and In 9 but not In 3, in which among about a thousand counted none had chromosome bridges at anaphase II.

It is of interest that the interchromosomal effect of inversions on crossing over is not apparent in maize-teosinte hybrids carrying only In 8 and In 9, but as soon as In 3 comes into the karyotype with these two inversions, the interchromosomal effect becomes distinct. It is not unlikely that this phenomenon is controlled by position effect as suggested by Steinberg and Fraser (1944) to explain a similar situation in *Drosophila*.

Table 1. Frequency and percentage of bridges and fragments observed at anaphases I and II in the sporocyte divisions of an F_1 plant of Wilbur's flint X Xochimilco teosinte in which In 3, In 8 and In 9 are present.

Division	Anaphase I						Anaphase II		
Class	O B O F	1 B 1 F F	1 B 2 F F	2 B 2 F F	O B 1 F	O B 2 F	O B	1 B	1 B 1 F F
Frequency	356	29	1	2	19	1	424	13	1*
Percentage	87.2	7.1	0.2	0.4	4.6	0.2	97.0	2.9	0.1

*unexpected

Y. C. Ting

9. Haploidy in the backcrossed progeny of a maize-Huixta teosinte hybrid.

Among the progenies of the third backcross to its maize parent (Wilbur's flint) of a maize-Huixta teosinte hybrid, a haploid plant was identified. It had 10 chromosomes instead of 20 as found in its sibs and it originated through gynogenesis. This plant was late in maturity, small in growth, and tillered profusely. During microsporogenesis, the sporocytes appeared much smaller than those of the diploid sibs. At early prophase, synizesis was always present. The identity of each univalent was then difficult to recognize. At pachytene, univalent chromosomes were extremely entangled, and they frequently folded back upon themselves to form nonhomologous associations. Pairing between heterologous chromosomes was rarely observed.

At metaphase I, practically all of the chromosomes appeared as univalents. Among a total of 308 randomly selected sporocytes, only eight had in addition to eight univalents, one bivalent possessing a chiasma-like appearance. Therefore, exchanges (or translocations) between two heterologous chromosomes are to be anticipated. At anaphase I, irregular chromosome distributions of various types were seen. Hence aneuploidy in the subsequent generation is expected to occur. A further investigation of this haploid plant is being carried on with the following objectives: (1) to study its derivatives by crossing to a standard

inbred strain of Wilbur's flint, (2) to compare the phenomenon of homozygosis with that of heterozygosis for teosinte chromosome segments by both selfing and crossing to its maize parent.

Y. C. Ting

10. Estimation of tripsacoid germplasm in teosinte and "Tripsacum" derivatives of maize.

In last year's News Letter, a new method for estimating teosinte and "Tripsacum" introgression into maize was described. This was based upon the comparative study of the cobs in a longitudinal section. While these studies are still in progress, another method has been found to be of some additional help. This involves crossing with Nobogame teosinte: (1) the original strain of A158, (2) strains of A158 modified by introducing teosinte chromosomes, (3) A158 strains modified by introducing extracted chromosomes from tripsacoid races of maize which are not in obvious contact with teosinte. The F_1 pistillate spikes have been studied for the following characteristics: (1) distichous versus polystichous arrangement, (2) single versus paired spikelets. The results for the first character which are based upon scores of 1-3 are shown in Table 1. The three grades are: 1 = distichous; 2 = intermediate; 3 = polystichous. In addition to this, those pistillate spikes having single spikelets are marked with one or two asterisks respectively depending upon whether less than or more than half the individuals of the F_1 population exhibit this feature. Absence of an asterisk indicates no single spikelets. Observations are based on 18-24 spikes from 9-12 plants.

Table 1. Results of crosses between modified and unmodified strains of A158 with Nobogame teosinte.

"Tripsacum" derivative X Nobogame teosinte		Teosinte derivative X Nobogame teosinte	
Country ¹	Average score	Derivative ²	Average score
Cuba	1.00*	Nobogame 4	1.0**
Honduras	1.07	Durango 1,9,7	1.0**
Nicaragua	1.18	Florida 4	1.0*
Bolivia	1.20	Florida 9	1.0*
Argentina	1.21	Florida 1,3 or 9	1.0*
Paraguay	1.27	Florida 3,4,9	1.0**
Brazil	1.33	Florida 3	1.1*
Mexico	1.43		
Control:			
A158 X Nobogame	2.6		2.6

¹Countries representing the source of races from which the chromosome with "Tripsacoid" effects has been extracted and introduced into A158.

²Varieties of teosinte representing the most likely source of chromosomes or chromosomal segments which have been introduced into A158.

It is obvious from the results set forth in Table 1 that the hybrids between A158 and Nobogame teosinte have a general tendency towards polystichous arrangement whereas those between modified derivatives and Nobogame show more tendency towards distichous arrangement. This demonstrates the fact that both types of derivatives carry concealed genes for distichous arrangement which is one of the distinguishing characteristics of teosinte and Tripsacum. Most of the "Tripsacum" derivatives, however, fail to show single spikelets. It is possible that Nobogame teosinte which is one of the most maize-like of the teosinte varietes, has failed to produce a threshold effect for this character. Crosses with Florida teosinte have been made during the past summer to see whether this teosinte would, in crosses, expose the hidden features in "Tripsacum" derivatives.

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1. Proembryo irradiation to produce blue-fluorescent and albino seedlings.

Maize plants were given approximately 750 r of gamma radiation (cobalt⁶⁰) at various times after pollination in order to produce chimeras in embryos, sectored for chlorophyll or carotenoid deficient phenotypes and normal tissue.

In one experiment the resulting embryos heterozygous for factors on chromosome 9 from the following cross were irradiated at various periods after pollination: wd* - C - sh - bz - wx/Yg₂ - C - Sh - Bz - wx (McClintock's rearranged chromosome*) X wx - Bf₁. The kernels from 10 ears of the treatment above were grown and the number of albino and blue fluorescent seedlings was observed. The loss of wild-type markers was independent in these two cases since these factors were on different homologous chromosomes. The results are given in Table 1.

Table 1. Frequency of seedling mutants resulting from irradiated proembryos.

Total seeds	Phenotype of seedlings				Period after fertilization of 750 r treatment
	wd +	wd Bf ₁	+ Bf ₁	+ + gl**	
723 (percent)	4 0.55	2 0.27	8 1.1	--	28 - 45 hours
596 (percent)	--	--	1 0.16	1** 0.16	52 - 68 hours

*See McClintock (1941) Genetics 26: 234-282 and (1944) Genetics 29: 478-502.

**Sectored seedling, 1/2 glossy and 1/2 non-glossy.

All of the irradiated embryos were heterozygous for Bf_1 , but only $1/2$ were heterozygous for wd . The observed loss of each (Bf_1 to wd is 11 to 6) was about as expected according to their position on the long and short arms of 9. It should be noted that the blue fluorescent plants, hemizygous for Bf_1 , were fluorescent in leaves 1, 2, 3 and 4 at the time plants were taken to the field. As indicated by E. G. Anderson (M.N.L. 33,6) Bf_1 Bf_1 plants fluoresce best in the first leaf and fluorescence greatly decreases in leaves that follow. Anthranilic acid appears to accumulate in greater amounts when Bf_1 is hemizygous.

Dale M. Steffensen

2. Patterns of sectoring in seedling with reference to early embryonic development.

From the studies of Stadler (1930, J. Hered. 21: 3-19) and Casper (M.N.L. 34,3) no sectored seedlings arose from ears irradiated during the first day after pollination. From the detailed account of early embryonic development presented by Randolph (1936, J. Agr. Res. 53: 881-916) one can deduce that sectored seedling could first be induced in the 6 or 8 celled embryo, which occurs at about 42 hours after pollination (maximum day temperature $27.5^\circ \pm 2.5^\circ$ C. and minimum night temperature $15^\circ \pm 2^\circ$ C.). A perfectly bilateral seedling could occur only when a transverse division had occurred (Fig. 4, G to N from Randolph, 1936), one cell carrying the dominant factor, the other deficient.

Preliminary observations of sectoring patterns have indicated that bilateral symmetry in seedlings is produced in proembryos irradiated 29 to 48 hours after fertilization. The plane of the leaf axis must be determined at this time because of the production of exactly bilaterally sectored seedlings having half green tissue on one side of the midribs and albino tissue on the other. During this 29 to 48 hour period, more completely albino seedlings were produced than seedlings with leaf area $1/2$, $1/3$, or $1/4$ sectored. In one seedling the leaves were completely albino but the coleoptile was half green and half albino indicating that the "anlagen" of the coleoptile is also determined during this period.

These experiments were not designed to study the early embryonic development of maize but were done to obtain sectored plants for cytoplasmic inheritance studies with chlorophyll and/or carotenoid mutants (wd , $w - 8624$, w_3 and $pastel - 8549$) in the hemizygotic state. Out of the 6,057 seeds planted, only one wd -sectored plant survived to maturity giving an ear with 3 inviable seeds. The method may be sound but will require larger populations and special care of sectored plants to assure seed set.

Dale M. Steffensen

3. Mosaic phenotypes from endosperm nuclei irradiated after fertilization.

The endosperm nuclei begin to divide before the embryo divides. In the period of irradiation (750 r of Cobalt-gamma rays) 28 to 48 hours after fertilization, treated endosperm nuclei heterozygous for the factors Sh sh sh , Bz bz bz and Pr pr pr gave at least 10% kernels with mosaic endosperm.

The range of the size of endosperm mosaics varies from 1/2 to 1/4 or smaller of the endosperm. Some of the bz-sh phenotype were of the usual breakage-fusion-bridge-cycle products. Endosperm nuclei treated between 52 to 68 hours after fertilization gave seed of which at least half contained one or more small mosaic losses. Quantitative studies could very well be done at this stage because of the relatively large number of sectors and ease of identification.

Dale M. Steffensen

4. Studies on induced non-disjunction.

In the studies where losses of the short arm of chromosome 9 have been followed in the endosperm and aleurone layers, it has always been assumed that losses of C - sh - bz - wx genes are due to a break between waxy and the centromere. A loss of the entire chromosome 9 would give the same result.

A homozygous 5-9 translocation (Anderson and Longley's 7205) carrying a dominant marker was used to test this latter possibility. The break in the long arm of 9 is attached to the segment of 5 containing Pr. Irradiated pollen was crossed to the recessive C - sh - bz - wx, pr thereby permitting one to detect loss on both the short and the long arm of 9. Roughly about 1/10 of the losses of Wx also lost Pr. As expected, it appears that chromosome breakage between wx and the centromere does explain the majority of losses. Coincident losses of Wx and Pr can be explained by two separate detachments (i.e. dicentric and centric rings) although a low frequency of complete chromosome loss cannot be ruled out.

Four different compounds were selected for tassel treatment because of their known properties and reactions with sulfhydryl groups. According to Mazia and others, sulfhydryl groups are reported to be involved with spindle function. The following compounds were used at the concentration indicated: 0.001 M $HgCl_2$, 0.005 or 0.0001 M. $CuSO_4$, 0.002 M iodoacetamide and 0.01% betamercaptoethanol. Solutions of these compounds were injected with a hypodermic syringe into the tassel well after meiosis had occurred with the idea of disturbing the second pollen grain division. The desired situation was to get 11 chromosomes in one sperm and 9 in the other.

Pollen from treated plants was then crossed to recessive testers as follows: C - sh - bz - wx, pr, y X c - Sh - Bz - Wx - Wc - Pr, Y (homozygous 5-9 translocation). Aberrant kernels possessing the recessive phenotype were selected from 4,593 seeds and planted in order to test the possibility that the plants were trisomic for chromosome 9. Root tips were collected from each of the 29 plants which resulted. Backcrosses were made to the recessive tester to detect any evidence of trisomic-type ratios and contamination. Of the 24 plants checked out completely, all were diploid as far as chromosome 9 was concerned. No clear cut case for non-disjunction giving rise to trisomic plants was found.

Dale M. Steffensen*

*Most of this work was done at Brookhaven National Laboratory under the auspices of the A.E.C.

5. Genetic study of B³ chromosome.A. Types of pollen produced by "3 3^B B³ B³" plant.

The A-B translocation involved here is TB-3a. Progenies from the cross $3a \underline{sh} \ 3a \underline{sh} \times \ 3a \underline{Sh} \ 3^B B^3 \ A \underline{Sh} \ B^3 \ A \underline{Sh}$ were analyzed in detail for the kernel phenotype, plant color, pollen abortion, and degree of glume clumping, and were crossed by and onto $3a \underline{sh} \ 3a \underline{sh}$ tester plants. The expected pollen types produced by the original male parent would be "3^B B³" and "3 B³". All possible plant types produced by these two kinds of pollen, taking account of nondisjunction of B³ at the 2nd microspore division and of crossing-over between the translocation point and A locus, were considered. Most of the plant types are distinguishable from each other in some way. The types of pollen and sperm involved in fertilization were determined on the basis of analysis of each individual plant from the parental cross.

About 55% of pollen functioning in the parental cross was the "3^B B³" type. Then 43%, (2% being unclassified), should have been the "3 B³" type. Among F₁ plants which came from colored, full kernels (Cl Sh), 75 individuals have the "3 3" genotype. They may have come from "3 B³" pollen. However, if "3" pollen had been produced by the "3 3^B B³ B³" plant, the resulting plants would also be "3 3". These "3 3" plants of different origins can not be distinguished.

However, Roman (Genetics 35:132, 1950) reported that the B⁴ chromosome in "4 B⁴" type pollen undergoes nondisjunction rarely if at all. If this is the case with B³ in "3 B³" type pollen, almost all "3 3" type plants must have come from "3" type pollen, except two colorless plants from colored Sh kernels. These two plants can be explained as a result of heterofertilization. The absence of "3 3 B³ B³" type plants is in agreement with Roman's statement.

As a result the percentages of the 3 types of pollen which functioned in the parental cross could be roughly estimated.

<u>Parental Cross</u>	<u>Type of Pollen Functioning</u>	<u>Percentage</u>
3 3 X 3 3 ^B B ³ B ³	3 ^B B ³	55%
	3 B ³	3%
	3	40%
	Unclassified	<u>2%</u>
		100%

B. Nondisjunction of the B³ chromosome in the "3 3^B B³ B³" plant.(1) Nondisjunction of B³ on the male side.a. Nondisjunction of B³ at meiosis.

The types of pollen produced by the "3 3^B B³ B³" plant turned out to be "3^B B³", "3 B³", and "3". To get the "3" type pollen, nondisjunction of the B³ chromosome must have taken place either at AI or at AII in meiosis. A cytological study of MII of microsporogenesis will clarify when this nondisjunction occurs. If any dyads show (10 + 12) chromosome distribution at MII, nondisjunction has

occurred at AI. In fact Blackwood confirmed cytologically that nondisjunction of B chromosomes occurs at AI (Heredity 10:345, 1956). Therefore it is conceivable that nondisjunction of B³ chromosomes also takes place at AI.

As a result of the meiotic nondisjunction of B³ in the "3 3^B B³ B³" plant, we have "3", "3^B B³ B³", "3^B", and "3 B³ B³" pollen. "3^B" is apparently aborted. "3^B B³ B³" pollen would produce a variety of plant types from crosses with normal egg parents. These plant types are as follows:

<u>Endosperm type</u>	<u>Plant type</u>
3 3 3 ^B B ³ B ³	3 3 ^B B ³ B ³
3 3 3 ^B B ³	3 3 ^B B ³ B ³ B ³
3 3 3 ^B B ³ B ³ B ³	3 3 ^B B ³
3 3 3 ^B B ³ B ³ B ³ B ³	3 3 ^B
3 3 3 ^B	3 3 ^B B ³ B ³ B ³ B ³

Three of the five plant types are distinguishable from plant types produced by "3^B B³" and "3 B³" pollen. These plant types were not found in the progeny.

"3 B³ B³" pollen is not expected to function because of its highly unbalanced chromosomal makeup which fails to compete with the normal or less unbalanced pollen in pollen tube growth.

b. Nondisjunction of B³ at the 1st microspore division.

If the pollen is the "3^B B³" type, nondisjunction of B³ at the 1st microspore division would produce either a deficient tube nucleus or a deficient generative nucleus. The deficient tube nucleus would prevent the pollen from functioning. The deficient generative nucleus will result in two hypoploid gametes which, after fertilization, produce hypoploid endosperm and hypoploid embryo. These were not found in the progeny studied.

If the pollen is the "3 B³" type, the consequence is different.

	<u>Tube nucleus</u>	<u>Generative nucleus</u>
After nondisjunction of B ³	3 B ³ B ³	3
	3	3 B ³ B ³

The plant type resulting from the generative nucleus of "3" type is not distinguishable from the type produced by "3" pollen. But the other type of generative nucleus results in a clearly distinguishable plant type; however, this type was not found in the progeny. Therefore it seems that the B³ chromosome disjoins normally at the 1st microspore division. Another support on this matter comes from Blackwood. She observed no nondisjunction of B chromosomes at the 1st microspore division (ibid., 1956).

Table 1. Types of Pollen Which Functioned in Parental Cross $3^a \underline{sh} \ 3^a \underline{sh} \times \ 3^a \underline{Sh} \ 3^B \ B^{3A} \underline{Sh} \ B^{3A} \underline{Sh}$

Kernel phenotype	Number of kernels	Number of plants analysed	Type of pollen										Unclassified	
			$3^B \ B^3$			$3 \ B^3$						3		
			Resulting plant type			Resulting plant type						Resulting plant type		
			$3^B \ B^3$	3^B	$3^B \ B^3 \ B^3$	$3 \ 3 \ B^3$		3 3		$3 \ 3 \ B^3 \ B^3$		3 3		
			colored plant	colorless plant	colored plant	colorless plant	colored plant	colorless plant	colored plant	colorless plant	colored plant	colorless plant		
Cl Sh	184	175	10	78		4		0	(2)	0	0	75		6
cl Sh	99	96	2	26			5	0	0	0	0		61	2
cl sh	98	72			72									0
Total	381	343	188			(11)						136		8
%			55%			(3%)						40%		2%

c. Nondisjunction of B^3 at the 2nd microspore division.

In this particular cross (cf. Table 1), the percentage of nondisjunction of B^3 in " $3^B B^3$ " pollen was 93%. If all colorless, shrunken kernels ($cl\ sh$), which were obviously hypoploid endosperm with hyperploid embryo, had germinated, the percentage would be higher. It would seem that hypoploid gametes tended to fertilize eggs more frequently, the apparent percentage being 59%. But if we take account of all $cl\ sh$ kernels, including ones which could not be analyzed because they failed to germinate, the percentage would approach 50%. This indicates random fertilization by both types of gametes, " 3^B " and " $3^B B^3 B^3$ ".

As discussed in section A, nondisjunction of B^3 in " $3 B^3$ " type pollen is not a common event. This can be checked genetically by planting colorless and colored kernels from $3^a 3^a \times 3^a 3^a B^3 A$ cross separately and scoring for the occurrence of colored and colorless plants respectively.

(2) Nondisjunction of B^3 on the female side.

a. Nondisjunction of B^3 at meiosis.

When $3^a \underline{Sh} 3^B B^3 A \underline{Sh} B^3 A \underline{Sh}$ plants were crossed by $3^a \underline{sh} 3^a \underline{sh}$ tester plants and the ears were analyzed, about 17.6% of kernels were $cl\ \underline{Sh}$. According to a rough calculation the expected frequency of $cl\ \underline{Sh}$ occurrence due to crossing-over within the T-A segment is about 14% at maximum. The excessive $cl\ \underline{Sh}$ kernels could be accounted for by meiotic nondisjunction of B^3 followed by formation of a "3" type megaspore. There is no way to tell the difference between nondisjunction of B^3 at AI and at AII except by cytological study. The actual ratio of expected megaspore types can be obtained by planting all kernels from the original cross and by classifying plants according to kernel phenotype, plant color, pollen abortion and degree of glume clumping.

b. Nondisjunction of B^3 at embryo sac formation.

If nondisjunction of B^3 takes place some time at embryo sac formation, the genotypes of polar nuclei and egg might be different. They can be determined by planting $cl\ \underline{Sh}$ and $Cl\ \underline{Sh}$ kernels separately and scoring for the occurrence of colored and colorless plants respectively.

Ikuko Mizukami

6. The effect of a-x deficiencies on crossing over in $T\ \beta\ a\ sh / N\ a-x$ plants.

Of the 109 alpha-bearing strands reported by Laughman (Mutation and Plant Breeding Symposium, 1961) among offspring of T-marked hemizygotes ($T\ \beta\ a\ \underline{Sh} / N\ a-x$), none carried the marker (N) proximal to the a-x deficiency. As was reported, their complete absence is somewhat surprising since they might be expected, at least occasionally, as a result of a coincidental exchange in the T- β segment.

Two obvious hypotheses to explain the rarity of this coincidental exchange are that the event giving rise to the nonrecombinant alpha has an interfering effect, or that the deficiency or its effect may extend well to the left of the A locus. The deficiencies a-x₁ and a-x₃ are of X-ray origin and are known to include the A locus and also to extend to the right beyond the Sh locus.

Data collected in this laboratory this past summer seem to bear on the hypotheses regarding the lack of N α Sh recombinants. Hemizygotes of the constitution T β α sh / N a-x were crossed by a homozygous colorless (a Sh / a Sh) pollen parent. From the F₁ ears, colored (β α sh / a Sh) and colorless (a Sh / N a-x) individuals were planted and determinations made on each individual plant for the presence or absence of aborted pollen. The presence of aborted pollen is typically associated with plants that are heterozygous for the translocation (T). In addition, individual suspect plants were either self-pollinated or crossed by a known pollen parent tester and at maturity the ears checked for the normal or aborted condition.

Preliminary results indicate the frequency of exchange in the T-β region is greatly reduced when the homologue is deficient. The normal frequency of recombination between T and β in T β α sh / N a Sh heterozygotes approximates 7.0 percent, whereas in the T β α sh / N a-x hemizygotes this frequency is reduced to 1.0 percent or less.

It appears from these data that the effect of the a-x deficiencies is a marked inhibition of exchange in the T-β segment of the hemizygote. Moreover, since the above experiment does not involve the isolation of the nonrecombinant alpha strand, the hypothesis of an interference due to this event seems unlikely as an explanation for the absence of N α Sh recombinants.

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1. Location of Rf_2 , a fertility-restoring gene for Texas sterile cytoplasm.

Two dominant complementary genes, Rf_1 and Rf_2 , are required for full restoration of male fertility in the presence of Texas sterile cytoplasm. Rf_1 has been shown to lie between d_1 and ts_1 on chromosome 3 (Duvick, Snyder, and Anderson, 1961, Genet. 46:1245-7)⁴

The following is a portion of the data obtained in recent X^2 tests involving Rf_2 and a series of chromosomal translocations:

Family	Translocation	FT*	FN	ST	SN	Total	P
60-3022	6-9d	1	16	12	5	34	<.01
61-21092	6-10(5519)	58	0	0	40	98	<.01

*F = fertile, S = sterile, T = translocation heterozygote, N = normal.

Although further tests are required, it appears that Rf_2 is located on the short arm of chromosome 6 at approximately 6S.75, the breakage point of T6-10(5519).

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1. Symbols for genes for resistance to rust, *Puccinia sorghi*, Schw.

The discovery of a second major gene locus for resistance to *P. sorghi* in inbred 25 from Australia (page 51, 1961 M.G.C.N.L.) raises the question of gene nomenclature. In keeping with genetic custom, new gene loci would be identified by a different subscript represented by different numbers; different alleles of the same locus would be identified by different small letters shown as superscripts. The symbol rp_2 has been used to identify the recessive gene in the sweet corn inbred 13-b for resistance to *P. sorghi* in Argentina (Page 39, 1948 M.G.C.N.L.). Therefore, the symbol Rp_3 is suggested for the locus in inbred 25 identified by rust culture 901aba. Since rp_2 and Rp_3 could easily be confused with Rp_2 present in inbred B38 and with Rp_3 present in inbred K148, especially in verbal communication, it is suggested that the symbol $\underline{R}p_1$ be used for the locus in inbreds GG208R,

B38, K148, etc. located near the end of chromosome 10 (Page 47, 1960 M.G.C.N.L.). The dominant genes for resistance in GG208R, B38, K148, Cuzco, B49, and P. I. 172332, previously designated as Rp^1 , Rp^2 , Rp^3 , Rp^4 , Rp^5 , and Rp^6 become Rp_1^a , Rp_1^b , Rp_1^c , Rp_1^d , Rp_1^e , and Rp_1^f , respectively.

A. L. Hooker

2. A gene for resistance to P. sorghi present in a resistant source from Mexico

Inheritance studies involving F_1 , F_2 , F_3 , and backcross progenies derived from a cross of a rust-resistant inbred M166 from Mexico with the susceptible inbreds B14 and R168 indicate that M166 contains a single dominant gene for resistance to P. sorghi. This is indicated by the following number of resistant, segregating, or susceptible progenies when tested with the rust culture 90laba.

Cross	Number of plants or progenies observed			Expected ratio	P value
	Res.	Seg.	Susc.		
(M166 x B14) F_2	121	0	30	3:0:1	0.10-0.20
(M166 x R168) F_2	89	0	33	3:0:1	0.50-0.70
(M166 x B14) x B14	82	0	67	1:0:1	0.20-0.30
(M166 x R168) x R168	73	0	68	1:0:1	0.50-0.70
(M166 x B14) x M166	141	0	0	all res.	
(M166 x R168) x M166	107	0	0	all res.	
(M166 x R168)	24	40	29	1:2:1	0.30-0.50

Hooker (Page 53, 1961 M.G.C.N.L.) has demonstrated that M185-1 has a single dominant gene for resistance which assort independently of the genes at the Rp_1 locus (Syn. A and B.Y. Dent) and that the single genes in M189 and M212 are either at or closely linked to the Rp_1 locus.

Inbred M166 was crossed with M185-1, M189, M212, and B.Y. Dent. These single crosses were advanced to the F_2 and crossed with the susceptible inbred Oh07K. The following data were obtained in greenhouse tests with rust culture 90laba which is avirulent to the resistant inbreds.

Cross	No. of plants observed		Expected ratio	P value
	Res.	Susc.		
(M166 x M189) F ₂	163	9	15:1	0.50-0.70
(M166 x M189) x Oh07K	413	144	3:1	0.50-0.70
(M166 x M185-1) F ₂	108	14	15:1	0.01-0.02
(M166 x M185-1) x Oh07K	408	148	3:1	0.30-0.50
(M166 x M212) F ₂	112	11	15:1	0.20-0.30
(M166 x M212) x Oh07K	352	144	3:1	0.02-0.05
(M166 x B.Y.Dent) F ₂	132	3	15:1	0.05-0.10
(M166 x B.Y.Dent) x Oh07K	432	141	3:1	0.80-0.90

These data indicate that the gene for rust resistance in M166 assort independently of genes at the Rp₁ locus and is at a different locus than the gene in M185-1. Work is in progress to determine the relationship of the genes in M185-1 and M166 to locus Rp₃. Both of these genes cannot be at locus Rp₃; therefore, at least one of these genes would be at a new locus.

W. L. Hagan

3. Reactions of certain corn relatives to Puccinia sorghi.

A number of corn relatives were tested for reaction to culture 901aba of Puccinia sorghi. Coix lacryma-jobi, Tripsacum lanceolatum, and 2 seedlings of T. dactyloides gave the chlorotic fleck reaction. Three other seedlings of T. dactyloides showed no symptoms.

Resistant and susceptible reactions similar to those of corn were expressed by annual teosinte (Euchlaena mexicana), but only chlorotic flecks were noted on perennial teosinte (E. perennis). Some progress has been made in transferring rust resistance from teosinte to corn. The high resistance of perennial teosinte (2n = 40) persisted after two backcrosses to tetraploid corn. A single dominant gene appeared to be involved. A lesser degree of resistance from annual teosinte (2n = 20) remained after two backcrosses to diploid corn.

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1. Crossing over in chromosome-3 as influenced by B-chromosomes.

Closely related lines of Black Mexican Sweet Corn with and without B-chromosomes were crossed to a chromosome-3 tester homozygous for gl₆, lg₂, a₁, et. Root-tips were obtained from the F₁ seedlings and the number of B-chromosomes possessed by each plant ascertained. The F₁ plants were then backcrossed to the chromosome-3 tester and the crossover frequencies between the gene markers determined.

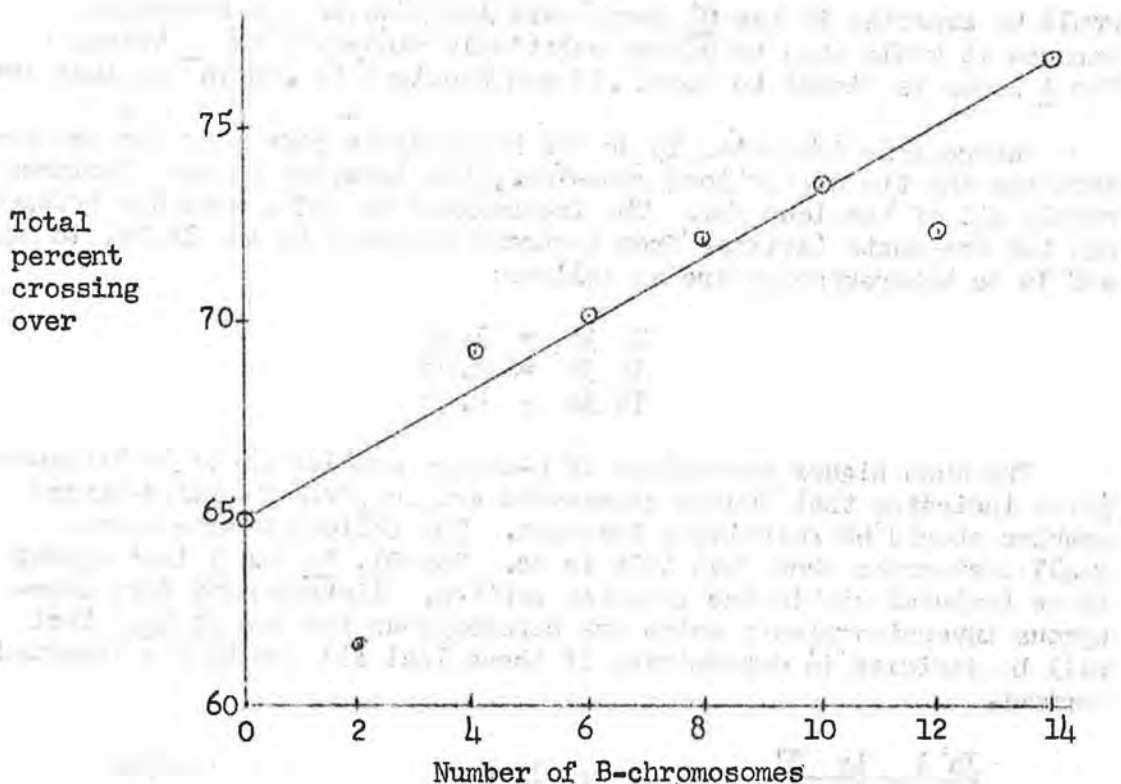
Frequencies of crossing over from a backcross of

G1	(1)	Lg	(2)	A	(3)	Et	♀	X	gl	lg	a	et	♂
gl		lg		a		et							
No. of B chromosomes		Total progeny		Percent crossing over									
				1		2		3		Total			
0		3111		21.8		31.5		11.6		64.9			
1-4		2300		24.5*		31.9		12.1		68.5**			
5-8		7153		26.5**		31.3		13.3*		71.1**			
9-12		3819		26.0**		33.8		13.5*		73.3**			
13-16		596		27.9**		34.6		14.4*		76.9**			

*, ** = Significantly different from 0 B-chromosome class at 5% and 1% level, respectively.

The data indicate a significant increase in crossing over due to the B-chromosomes in the gl-lg and the a-et regions. Crossing over in the lg-a region is increased but the amount is not significant. The total percent crossing over between gl and et indicates a significant increase in the presence of the B's and a graphic plot of the data suggests that the effect of the B-chromosomes is additive.

Studies are being made to determine the interaction, if any, of B-chromosomes and knobs on crossover frequencies.



George P. Hanson

2. Further studies with heterozygous inversions in chromosome 3.

The paracentric inversion In 3b has breakpoints at positions .25 and .75 in the long arm of chromosome 3. Backcross data from In 3b heterozygotes presented in the 1956 News Letter gave the following recombination values:

$G1_6-Lg_2$	0.57%
$G1_6-A_1$	9.7%
Lg_2-A_1	9.6%
A_1-Et	17.7%

It is apparent that the A locus is distal to point .75 in the long arm, but the close linkage of G1 with Lg could arise if both loci were included in the inversion or if G1 were in the proximal uninverted segment. In order to delimit more precisely the cytological location of G1, plants homozygous for In 3b and heterozygous for the G1 and A loci were testcrossed as shown below:

$\frac{g1}{G1}$ In A	X	gl a	gl A	G1 a	gl a	G1 A	$\Sigma = 665$
G1 In a			193	165	127	180	

The 46.2% of recombination between G1 and A indicates that G1 is in the proximal uninverted segment. A much lower recombination value

would be expected if the G1 locus were included in the inversion because it would then be placed relatively closer to the A locus. The A locus is distal to point .75 and proximal to .95 in the long arm.

Paracentric inversion In 3c has breakpoints very near the centromere and the tip of the long arm--i.e., the inverted segment includes nearly all of the long arm. The frequencies of PMC's with two bridges and two fragments (arising from 4-strand doubles) in the In 3a, In 3b, and In 3c heterozygotes are as follows:

In 3a = 1.2%
In 3b = 1.0%
In 3c = 8.5%

The much higher percentage of 4-strand doubles in In 3c heterozygotes indicates that double crossovers arising from 2- and 3-strand doubles should be relatively frequent. The following data from a small test-cross show that this is so. The G1, Lg and A loci appear to be included within the inverted section. Linkage data from homozygous inversion plants which are heterozygous for the G1 Lg A loci will be decisive in determining if these loci all are in the inverted segment.

		<u>In A</u>			<u>Lg</u>			<u>G1</u>			X				$\Sigma = 526$	
		<u>N</u>	<u>gl</u>	<u>lg</u>	<u>gl</u>	<u>lg</u>	<u>a</u>	<u>gl</u>	<u>lg</u>	<u>a</u>						
(0)	(0)	(1-2)	(1-2)	(1-3)	(1-3)	(1-4)	(1-4)	(2-3)	(2-3)	(2-4)	(2-4)	(3-4)	(3-4)			
In	N	In	N	N	In	N	In	In	N	In	N	In	N			
<u>G1</u>	<u>gl</u>	<u>G1</u>	<u>gl</u>	<u>gl</u>	<u>G1</u>	<u>G1</u>	<u>gl</u>	<u>G1</u>	<u>gl</u>	<u>gl</u>	<u>G1</u>	<u>gl</u>	<u>G1</u>			
<u>Lg</u>	<u>lg</u>	<u>Lg</u>	<u>lg</u>	<u>Lg</u>	<u>lg</u>	<u>Lg</u>	<u>lg</u>	<u>lg</u>	<u>Lg</u>	<u>lg</u>	<u>Lg</u>	<u>lg</u>	<u>Lg</u>			
<u>A</u>	<u>a</u>	<u>a</u>	<u>A</u>	<u>A</u>	<u>a</u>	<u>A</u>	<u>a</u>	<u>A</u>	<u>a</u>	<u>A</u>	<u>a</u>	<u>A</u>	<u>a</u>			
268	200	3	11	1	3	1	4	14	11	8	0	2	0			

M. M. Rhoades

3. Linkage studies with chromosome 3.

Two backcrosses were made of F_1 's segregating for lg₂ na₁ a₁. F_1 plants of family 24575 had knobless chromosomes 3 while F_1 plants of family 23529 were heterozygous for a large knob at 3L.6. The two families are not closely related; one of the parents of 24575 was inbred KYS which is not present in the 23529 material. The backcross data are presented below. The order of the genes is lg-na-a. Previous studies with heterozygous knobs have shown that crossing over is reduced in regions near the knob. The finding of a higher Lg-Na value and a lower Na-A value in K3/k3 plants might be taken to indicate that the knob is distal to na. However, comparisons of the two sets of data are not valid because of the different genetic backgrounds.

Constitution of backcrossed families	Lg Na A	Lg Na a	Lg na A	Lg na a	lg Na A	lg Na a	lg na A	lg na a	Σ	% Lg-Na	% Na-A
24575	333	163	2	6	9	0	145	278	936	1.8	33.1
$\frac{\text{Lg Na A}}{\text{lg na a}}$ - $\frac{\text{k}}{\text{K}}$	(0)	(2)	(1-2)	(1)	(1)	(1-2)	(2)	(0)			
23529	43	17	436	1061	1240	486	11	31	3325	3.1	28.6
$\frac{\text{Lg na a}}{\text{lg Na A}}$ - $\frac{\text{k}}{\text{K}}$	(1)	(1-2)	(2)	(0)	(0)	(2)	(1-2)	(1)			

Since the knob was thought to be close to Lg, sporocytes were collected from crossovers between Lg and Na (in the progeny of the 23529 backcross) and the chromosomes examined for presence or absence of the knob. Twelve Lg Na A plants had the knob as did four Lg Na a plants. One Lg Na a plant was knobless. Two lg na a plants and one lg na A plant had knobless chromosomes 3. The knob is closer to Na than to Lg; it cannot be proximal to Lg, but it is uncertain whether it is proximal or distal to Na.

Studies on the effect of knobs on crossing over have been extended to include plants homozygous for a large knob at position 3L.6. As reported earlier (MNL No. 31, 1957) recombination in the Lg-A region, which includes the knob, is significantly less in plants heterozygous for the knob than in plants which have homozygous knobless chromosomes 3, but the recombination values in plants with two knobbed chromosomes 3 remained to be determined. The following data were obtained from three classes of sib plants all of which, however, are heterozygous for abnormal 10.

K10 k10	Lg K A/ lg K a	-- 42.7% recomb.	Lg-A	Σ = 2129
K10 k10	Lg K A/ lg k a	-- 30.5%	" "	Σ = 3003
K10 k10	Lg k A/ lg k a	-- 34.1%	" "	Σ = 973

These data exhibit, although to a lesser degree, the previously reported reduction in heterozygous knobbed plants as compared to knobless plants, but the somewhat unexpected feature is the high crossover value found in homozygous knobbed plants. One can only speculate about the cause of this increase. It is true that a knobbed chromosome is longer, by the length of the knob, than is a knobless chromosome, but for this to have significance would seemingly require that crossovers occur in the heterochromatin comprising the knob. Perhaps a more likely hypothesis is that the fusion of knobs known to occur during interphase leads to earlier and possibly more intimate pairing in meiosis of the homologous regions flanking the knob and hence to an increase in crossing over.

Data presented in the 1957 News Letter suggested that the linear order, proceeding from the centromere, was Rg Cl₆ Lg₂ A₁, but the 4-point backcross population consisted of only 341 individuals. That this order is indeed correct is shown by the following test-cross data:

		$\frac{Rg \ gl \ lg \ a}{rg \ + \ + \ +}$				X	rg gl lg a				$\Sigma = 1165$			
(0)	(0)	(1)	(1)	(2)	(2)	(3)	(3)	(1-3)	(1-3)	(2-3)	(2-3)	(1-2-3)	(1-2-3)	
Rg	rg	Rg	rg	Rg	rg	Rg	rg	Rg	rg	Rg	rg	Rg	rg	
gl	+	+	gl	gl	+	gl	+	+	gl	gl	+	+	gl	
lg	+	+	lg	+	lg	lg	+	+	lg	+	lg	lg	+	
a	+	+	a	+	a	+	a	a	+	a	+	+	a	
256	270	4	7	89	97	196	162	2	4	38	39	1	0	

$$Rg-gl = 18 \div 1165 = 1.5\%$$

$$gl-lg = 264 \div 1165 = 22.7\%$$

$$lg-a = 442 \div 1165 = 37.9\%$$

Order is Rg gl lg a.

These data together with those from the lg-na-a backcross and from linkage data previously reported give the following linear order of genes in the long arm of chromosome 3, which is one of the best-marked arms of the maize chromosomes.

Rg gl₆ ts₄ lg₂ na₁ a₁ sh₂ et ga₇

M. M. Rhoades
Ellen Dempsey

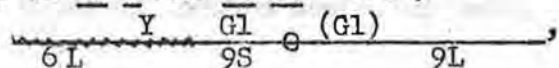
4. Tassels with 4N sectors.

In the course of pollen examination in a backcross population segregating for a translocation, it was noted that many tassels were sectored for two types of anthers. One type had small grains of the usual size and the other had larger grains. In plants expected to be heterozygous for the translocation, the first type of anther showed 50% abortion but the second type had mostly normal grains. Three plants were used as males in crosses to a 4N pr stock. Two of these carried the translocation and the other had normal chromosomes. The resulting ears had many plump seeds as well as some shrivelled seed. One ear had all Pr seed while the others were segregating Pr and pr. Ratios of 62 Pr: 15 pr and 91 Pr: 24 pr indicate the pr allele was in duplex as would be expected if doubling had occurred in a Pr/pr plant. Apparently the tassels contained tetraploid sectors. No such sectors were found in related material the previous year. The population in question was located downhill from an experiment involving treatment of seeds with various chemical mutagens. It is possible that washing of these chemicals affected the developing tassels and caused somatic doubling.

Ellen Dempsey

5. Linkage of Gl₁₅ and Y₁ in homozygous T6-9b.

A population of 1070 plants from a backcross of $\frac{y \ T \ Gl \ wx \ c}{Y \ T \ gl \ Wx \ C}$ was found to give 34.3% recombination for $\frac{C-Wx}{Y-Gl}$, 7.3% $\frac{Y-Gl}{C-Wx}$ recombination and independence of $\frac{Wx-Y}{C-Wx}$ and $\frac{Wx-Gl}{Y-Gl}$. The 9th chromosome can thus be represented



with Y in 6L and G1 close to the centromere either in 9S or 9L.

Ellen Dempsey

6. Linkage of oy.

The data listed below come from crosses of oy R k10/Oy r K10 females with oy R k10 males. Two types of kernels were produced, R R R and r r R. The r r R class is more frequent because of preferential segregation. There is no evidence of preferential segregation of oy or of linkage of oy and R.

			<u>R R R</u>		<u>R r r</u>	
22843	X	23063	<u>Oy</u>	<u>oy</u>	<u>Oy</u>	<u>oy</u>
			138	140	399	407

Because of the negative results obtained above, a further test of the location of oy on chromosome 10 was made. Plants trisomic for chromosome 10 were crossed to an oy stock and the trisomic F_1 's were used as male parents in the backcross to oy. Five different male parents gave ratios of green to oil yellow as follows:

	<u>Oy</u>		<u>oy</u>
24610-4	59		21
24610-11	56		22
24610-13	190		110
24612-12	42		23
24612-15	<u>70</u>		<u>27</u>
	417		203

The total of 417 green to 203 oil yellow indicates that oy is located on chromosome 10, as was reported by E. G. Anderson (MNL 25, 1951). Although abnormal 10 is present in the trisomic stock, no distortion of ratios is expected since male gametes were tested. A stock of du, which is 20 units proximal to R, was obtained from H. H. Kramer and will be crossed to oy for more precise location. The information given here indicates that oy is either in 10S or is close to the centromere in 10L.

Ellen Dempsey

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Department of Genetics and Selection

1. A study of the local and regional maize populations (*Zea mays* L.).

Maize in the zones of its original cultivation (for instance in the USA) became, in a relatively short time, the subject-matter of detailed studies in the most varied directions. These studies were based on collections gathered with ant-like zeal by research workers. In the course of time these investigations of the collections terminated (in our opinion) materially, and today the whole extent of maize has, on principle, been exhausted, studied and evaluated, the prospective types being in a considerable measure utilized in the selection practice.

In other zones (for instance in Czechoslovakia) this is far from the case. Maize in Central Europe was late in becoming an article of economic utilization, which also necessarily resulted in detailed scientific investigation. The inevitable consequence of this state of things was, among others, that the local and regional maize populations retained for a long time their positions in farming practice. The situation was further stressed by the rich geographical articulation of Central and Southern Europe and the historically different (often contrasting or crossing) articulation of trade routes that were likewise of great assistance in the historical dissemination of agricultural produce in Europe. These conditions (and probably others as well) resulted in different and genetically immensely valuable populations. These began to be utilized and improved in the selection practice, but the process of collecting, investigation and utilization has not by far been terminated.

In Czechoslovakia great attention is being paid to the questions of collecting, study and utilization of local and regional maize populations. In area (127.858 km²) Czechoslovakia does not range among the large countries, but her position and geographical articulation made it possible to originate here interesting populations. A certain contrariety is reflected in the fact that Czechoslovakia has always belonged to the progressive countries and has left an influence on the preservation of these populations. Another important factor was the establishment of collective large-scale agricultural units which made possible a complete introduction of the present-time technique and technology, organization of work, and the use of the most effective variations and hybrids in all production areas. There arose in dead earnest the question of an accelerated termination of maize collection. The Staff of the Department of Genetics and Selection, assisted by the pupils of agricultural and other schools in the most varied degree of instruction, began to collect some time ago, and has, at the present time terminated the detailed collection of the local and regional populations. On the whole 536 sites in Czechoslovakia have been covered. Special attention was paid to submontane and montane valleys (because of the cold-resistant and early-crop types). The relatively dense covering of the whole area resulted in duplications, in some instances, but in a work like this, this is immaterial. Representatives of the following varieties have been secured: *Zea mays* L.: v. *Indurata*, v. *Indentata*, v. *Everta*, v. *Saccharata*. The stage of collecting has thus been terminated. Now the main attention is being devoted to substantial studies and

evaluation, the most valuable material being placed at the disposal of workers at the Selection Stations. It is not possible in this short preliminary report to present an analysis of the results obtained. Some interesting facts can, however, be mentioned.

1. A valuable starting material for special selection has been gained. This is characterized by a considerable cold-resistance, a relatively quick start in the first stages of growth (our regions are distinctive for relatively cold springs), and relatively short vegetation periods.

2. In the genetic studies of the secured materials and chiefly in the genetic disintegration due to selfpollination we frequently met with the type-lines (a part of the material has been worked up into $S_6 - S_8$) which, as regards their taxonomico-anatomical structure, are very much like the respective classical, present-day lines in the USA.

3. The material obtained from the more eastern and central zones is frequently characterized by a fairly high farmability. Populations obtained from central (in which case there is a clear conflict of directions), eastern, northern and southern zones are characterized by lower farmability. From this one may infer that maize populations spread to this country from the more western countries (chiefly via western Germany) already in a certain degree of improvement and selection. Populations that had reached this country chiefly from the Balkans were not to such a high degree subject to human selection and have retained great variety and genetic width (this may have been caused by a greater geographic articulation of the places of transition and of those of cultivation). Some results point out to the basic directions of advance into this country even with regard to the individual zones.

4. As a complement to these studies populations from other countries of Europe are being collected. Also from this fairly rich material a whole series of prospective types for detailed study and utilization in special selection has been evaluated. These materials are also subject-matter for investigation.

Luděk Říman

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1. The mutability of the components of the E_n system.

The varied forms of E_n : E_n (Enhancer) is necessary for the mutability of the $a_1^{m(r)}$ allele. In the absence of E_n this allele is colorless and indistinguishable from other colorless alleles. The pattern of mutability expression is a result of rate (low and high) and time (early and late) of mutation events. Given a common $a_1^{m(r)}$ allele, different E_n can cause a predictably different mutable expression. In addition, each of the E_n isolates shows somatic changes.

The varied forms of "I": In the E_n system of mutability, the element suppressing the action of the locus has been designated "I" (Peterson, Genetics 46). When adjacent to the locus, the action of the

locus is suppressed. "I" is found at the A_1 and at the Pg locus. \underline{En} is specific for the "I" component and the introduction of \underline{En} results in mutability. In some cases, however, differing patterns of mutability are considered to be due to differences in "I". By crossing a single specific \underline{En} to an array of independently derived $a_1^{m(r)}$ alleles different expressions are observed. This indicates that the change must be in "I" and additional studies are being carried on to determine the nature of this change.

Thus, the final pattern is dependent on the particular \underline{En} as well as the particular "I" and/or the interactions between them. This is unlike the mutable pericarp locus where pattern differences result from the varied number of $\underline{tr-Mp}$ present.

Peter A. Peterson

2. A dominant mutable.

Among a group of r mutants originating from standard R , there occurred a seedling mutable characterized by dark stripes on a virescent-like background. Outcrosses of this mutant to green plants of Dr. Brink's color converted W-22 strains (a strain which has not given rise to any seedling mutants in our cultures) yielded progeny, 1/2 of which were similar in expression to this same mutable. This type of mutable has not previously appeared among the numerous mutables studied in our cultures. It would seem, therefore, that this represents the origin of a dominant mutable allele.

Peter A. Peterson

3. Pales at the a_1 locus.

Pales, both stable and mutable, arise from certain a^m alleles. They arise from the same autonomous alleles that give rise to different pattern types in the presence of \underline{En} . Stable pales are similar to $a^{m(mr)}$ in that they do not respond to independent \underline{En} . Neither do the mutable pales show any response to \underline{En} . The individual isolates of the stable pales show a wide range of expression from those displaying only a slight amount of color to those possessing deep pale color.

Peter A. Peterson

4. Knob and centromere associations of non-homologous maize chromosomes at pachytene.

This report is an extension of previous studies on the non-homologous association of knobs and centromeres (S. R. Peterson, M.S. Thesis--Univ. of Ill.; Gurgel MGCNL 30 and 31):

These studies were undertaken with stocks possessing 8 and 12 knobs in the hemizygous condition and were derived from a standard genetic line and maize chapolote, respectively, crossed with Tama knobless flint. The table below shows that more knob association and more multiple association occur in the higher knobbed family than in the lower knobbed families.

Family	# of knobs	% of cells having knob association	% of associations which were of 3 or more knobs
848	2	4.99	----
844	8	43.14	5.15
862	12	100.00	50.64

Knob association appears to be related to knob size--larger knobs tend to associate more frequently than smaller knobs. This agrees with earlier reports of Longley, Peterson and Gurgel.

There is also a relationship between knob association and the distance of the knob to the end of the chromosome arm; those knobs farther from the end of the arm appear in association more frequently than those closer to the end of the chromosome arm. From multiple regression analysis however, it was determined that knob size is more influential in associations than is knob position.

More chromosomes appear in centromere association and more associations of 3 centromeres occur in the 12 knob family than the 8 knob family.

The frequency of centromere association in this material appears unrelated to chromosome length. This is inconsistent with the observations in the KYS material (Peterson and Gurgel).

Some knob association persisted from pachytene to metaphase I. This was verified from the observation that more associations were seen at the various stages in the 12 knob strain than in the cells containing 2 knobs which agrees with the analysis of association at pachytene. The 8 knob cells gave intermediate values at all stages except diakinesis where fewer bivalents were associated than were found in the 2 knob cells.

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1. Tests of white-seeded lines of corn of undetermined genotype for allelism with y_1 .

Results of suppressor and allele studies with the white-albino (white endosperm, albino seedlings) mutants have indicated that the genes involved might have a complex structure consisting of two portions, one responsible for carotenoid synthesis in the endosperm and the other controlling carotenoid production in the seedling. Studies with some of these mutants have indicated that the two parts of the complex can be modified independently by suppressor genes or mutation. (Maize Genetics Cooperation News Letter 34:69-70, 1960). The discovery of the y_1 alleles, pas_{8519} , and w_{mut} (white endosperm, pale green seedling) suggests that y_1 might be the result of a mutation at a white-albino locus involving just the endosperm element of the complex. If this is the origin of y_1 , the gene responsible for most of our white

endosperm lines of corn, it is possible that a mutation involving just the endosperm portion of the complex of one of the twelve known white-albino loci may have given rise to white endosperm lines of corn that are non-allelic to y_1 . To search for such genes, 591 white endosperm accessions were obtained from the plant introduction station at Ames; these were grown along with two white endosperm mutants obtained from Mr. Kermicle and three from Dr. Bianchi, giving a total of 596 lines. An attempt was made to self pollinate two plants from each of these lines and at the same time outcross them to known y_1y_1 stocks. Any plant that was white endosperm because of a mutation of some locus other than y_1 should give yellow seeds in this outcross. Outcross tests were successful for 539 of the unknowns. Table 1 summarizes the results for the crosses where something other than white seeds was observed in the outcrosses.

Table 1

Class	Results			Source of accession
	Outcross to y_1 tester	Self pollination	Number of plants tested	
1	segregating yellow	segregating for recessive white	18	
2	segregating yellow	segregating for dominant white	7	
3	segregating yellow	homozygous white	2	1 Ethiopia 1 U. S. S. R.
4	homozygous yellow	homozygous very pale yellow*	8	2 Turkey 1 Ethiopia 1 New York State 2 Washington State 1 (Mutant furnished by Mr. Kermicle, Wisc., 2 tests made)
5	homozygous yellow	segregating very pale yellow*	3	2 Ethiopia 1 Paraguay
6	homozygous very pale yellow*	homozygous white	1	
7	heterozygous very pale yellow *	homozygous white	16	
8	homozygous very pale yellow *	segregating very pale yellow *	8	

Table 1, continued

Class	Results			Source of accession
	Outcross to Y_1 tester	Self Pollination	Number of plants tested	
9	heterozygous very pale yellow*	segregating very pale yellow*	12	
10	homozygous very pale yellow*	no test	2	
11	heterozygous yellow	red pericarp	2	
12	homozygous yellow	homozygous yellow	6	

*There is extremely little yellow pigment in the seeds classified as very pale yellow in this report. Such very pale yellow seeds have been frequently observed on ears that are otherwise $Y_1Y_1Y_1$ and they are probably due to modifier genes. The amount of pigment is considerably less than that found in seeds of the $Y_1Y_1Y_1$ genotype. In selecting Y_1 testers for these crosses, only pure white seeds were planted. However, because of a shortage of good Y_1Y_1 stocks, some white seeds were selected for use from ears that were segregating for very pale yellow seeds.

Class one probably represents cases where heterofertilization had resulted in white seeds being planted that carried Y_1Y_1 embryos. Plants of class two were heterozygous for a dominant white gene. Class three could represent cases where the unknown was $Y_1Y_1 Y_xY_x$. The plants of classes four and five could represent instances where the unknowns were $Y_1Y_1 Y_xY_x$. The presences of a slight tinge of yellow in the outcross seeds (very pale yellow) may or may not be significant since Y_1 in some backgrounds also is very pale yellow. (See footnote to Table 1.) Classes 6-10 represent crosses where very pale yellow seeds are present. Endosperm color of the selfed plants of class 11 could not be determined with certainty because of the presence of red pericarp color. Class 12 consists of instances where there was some doubt as to whether the seeds of the accessions planted were white. In all these cases the planted seeds had considerable yellow pigment but compared to other seeds on the ear they were definitely pale.

The 13 plants from classes 3, 4 and 5 are the ones most likely to be carrying a new white endosperm gene and these will be tested further. The significance of very pale yellow seeds in some plants from classes 6-10 will be investigated further although there is very little likelihood that a major white endosperm gene is involved.

If on further testing, some of the promising lines do turn out to

possess a new white endosperm gene, these will then be tested against our white-albino mutants to determine if any of these loci are involved.

Donald S. Robertson

2. Chlorophyll, carotene and xanthophyll production in pastel-8549, pastel-4889 and pastel-8686 grown at high and low temperatures.

Chlorophyll, carotene and xanthophyll levels were determined for the following white endosperm-pastel (pale green) mutants and their F_1 's with available albino alleles after growing under a light intensity of 1400 foot candles and at temperatures of 22° C and 37° C.

<u>Mutant</u>	<u>Chromosome</u>
pastel-8549	6 (y_1 allele)
pastel-4889	7
vp_9 (albino allele) / pastel-4889	
pastel-8686	3
w_3 (albino allele) / pastel-8686	

Homozygous pastel and F_1 's with albino alleles are possible for the latter two loci. The pollen parents for the F_1 's were selfed and served as the source of the homozygous pastels. For each mutant and F_1 tested, seed was separated on the basis of endosperm color into normal (yellow) and mutant (white) classes and a sample of each was planted in rows in sand and grown under the above conditions of light and temperature. Plants were grown for 7 days at 37° C and 13 days at 22° C before harvesting. The methods of extracting the pigments and determining concentrations are described by Robertson and Anderson (Temperature sensitive alleles of the y_1 locus in maize. Jour. of Hered. 52:53-60. 1961). The pigment concentrations for each mutant are given in Table 1 and the percentage of pigments in the mutant as compared to that in the normal siblings at the two temperatures is given in Table 2.

Table 1. Chlorophyll, carotene and xanthophyll levels for normal and pastel seedlings. (mg/gfw is milligrams per gram fresh weight.)

Mutant	temp. °C.	Normal			Mutant		
		chlorophyll mg/gfw	carotene mg/gfw	xanthophyll mg/gfw	chlorophyll mg/gfw	carotene mg/gfw	xantho. mg/gfw
pas-8549	37	2.373	.0777	.0621	.329	.0517	.0140
	22	2.551	.0531	.0702	1.737	.0489	.0453
pas-4889	37	2.708	.0755	.0731	1.189	.0391	.0243
	22	2.746	.0573	.0746	.536	.0129	.0297
vp_9 F_1 / pas-4889	37	2.568	.0873	.0586	.429	.0121	.0254
	22	2.575	.0809	.0960	.203	.0082	.0147
pas-8686	37	2.860	.0787	.0407	1.704	.0483	.0589
	22	2.477	.0658	.0407	.275	.0052	.0183
w_3 F_1 / pas-8686	37	2.411	.0903	.0670	.550	.0128	.0306
	22	3.546	.1105	.0780	.100	.0024	.0100

Table 2. The $\frac{\text{mutant}}{\text{normal}}$ values for chlorophyll, carotene and xanthophyll.

Mutant or cross	Temp. in °C.	% mutant normal	% mutant normal	% mutant normal	% mutant normal
		Chlorophyll	Carotene	Xanthophyll	Total carotenoid
Pastel 8549	22°	68.1	91.9	34.3	59.1
	37°	13.9	66.7	22.6	47.1
Pastel 4889	22°	19.5	17.2	50.9	32.1
	37°	43.9	51.7	31.6	42.6
vp9/Pastel 4889	22°	7.9	10.1	15.3	12.9
	37°	16.7	13.9	54.5	28.0
Pastel 8686	22°	11.1	7.8	44.9	22.0
	37°	59.6	61.4	164.9	93.7
w3/Pastel 8686	22°	2.8	2.3	12.8	6.6
	37°	22.8	14.1	45.7	27.6

In these three mutants the concentrations of all three of the chloroplast pigments have been affected. The pigment levels of pastel more closely approximate those of normals when grown at 22°C than 8549 at 37°C. This is in agreement with previous experiments with this mutant grown at 115 foot candles (Robertson and Anderson, Temperature sensitive alleles of the y_1 locus in maize. Jour. of Hered. 52:53-60. 1961). Pastel 8686 and pastel 4889 behave in an opposite manner with more normal appearing phenotypes observed at high temperature than low temperatures. Of the latter two mutants, pastel 8686 more closely approximates normality than does pastel 4889.

In comparing the results of each homozygous pastel with those of the F_1 between that pastel and the appropriate albino, it can be seen that neither the albino or pastel alleles of the two loci show complete dominance with respect to the other under these experimental conditions.

Lou Betty Richardson

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1. Defective endosperm factors in maize teosinte derivatives*.

Other allelism tests have been carried out among stocks possessing de^t factors. Allelism has been confirmed for de^{t3} and de^{t5} , and established for the latter and de^{t16} .

A. Bianchi

*Work financially supported by the Rockefeller Foundation, New York.

2. A new type of chromosome 9 in Italian maize*.

On the long arm of chromosome 9 a characteristic long shaped knob has been repeatedly found, in the position which has been reported as typical for the presence, in some cases, of a prominent chromomere. Such a type has been detected only in "Nostrano dell'Isola," a variety of special interest from the point of view of its origin and evolution, among Italian maizes, according to an oral communication of Drs. Brandolini and Wellhausen.

The finding is a part of the cytological survey of the pachytene chromosomes of Italian maize, which is being undertaken in cooperation with the Maize Breeding Station of Bergamo.

A. Bianchi

3. Mendelian characters in Italian maize*.

Self pollination has been carried out in plants of about 90 samples of Italian maize kindly provided by "Stazione di Maiscoltura di Bergamo."

The following mutants have been obtained in a total of 547 selfed ears:

Character	No. of cases exhibiting a ratio of	
	3:1	15:1
Defective seed	18	
Small seed	1	
Amylaceous seed	1	
Lemon endosperm	1	
Sugary endosperm	1	
Albino seedling	5	2
Dwarf seedling	6	
Seedling color (booster, etc.)	28	3
Luteus seedling	3	
Yellow green seedling	8	
Pale green seedling	8	2
Fine stripe seedling	5	
Glossy seedling	18	2
Abnormal growth	1	2
Virescent	10	3
Yellow stripe	1	1
Albescent		1
Japonica seedling	1	1
Virescent fine stripe	2	
Green mottling seedling	1	

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M. Pozzi

*Work financially supported by the Rockefeller Foundation, New York.

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1. Golden-2.

The location of golden-2 still seems to be in doubt if my survey of the News Letters is complete. In recent years it has been included in the long arm of the seventh linkage group near Bn, twenty units to the right of ij, in a number of publications, apparently on the basis of the following data from Sprague (N. L. 14, 1940).

g₂ ij RS 310 118 101 3 532 20% recombination

Brink and Arney in 1942 (N.L. 16, p. 34) report linkage of g₂ with T3-7b (3S.90; 7L.03) of 11.3% while earlier in the 1937 News Letter (p. 14), Brink had reported g₂ and d₁ linkage indicating a chromosome 3 location. I have some data to report which do nothing to resolve the matter.

Genes X Y	Phase	X Y	X y	x Y	x y	Total	Recombination
<u>Chrom. 3</u>							
<u>G₂ Cr</u>	CB	93	97	86	80	356	ca 51%
<u>Chrom. 7</u>							
<u>G₂ Pn</u>	CB	72	77	92	99	340	ca 50%
<u>G₂ Pn</u>	CS	126	40	96	40	302	ca 42%
<u>G₂ Bd</u>	RS	168	54	62	18	302	ca 49%

Only the coupling-self (CS) of G₂ Pn shows a significant deviation from the expected 1:1 ratio of independent gene segregation. Since I find Papyrescent (Pn) somewhat difficult to classify in certain cultures, this family may not be indicative of linkage.

Robert I. Brawn

2. Golden-4.

A variety of observations suggest that the stock of golden-4 carried by the Coop is not really golden-4, but more likely golden-1. The F₁ of g₁ x g₄ is golden in colour. In addition a supposedly 3-point linkage test with wx and bm₄ on chromosome 9 showed no significant deviation from a 1:1:1:1:1:1:1:1 ratio indicating independent assortment of golden with both wx and bm₄. The previously indicated position of g₄ is between wx and bm₄.

F ₁	Parental	Region 1	Region 2	Region 1-2	Total				
<u>+ + +</u>	74	67	77	77	70	61	62	50	538
<u>wx g₄ bm₄</u>	141	154	131	112	26.2%	28.6%	24.3%	20.8%	

Robert I. Brawn

3. Origin of inbred Pa703Y.

The Pennsylvania station has isolated a colourless pericarp, red cob subline (Pa703Y) of the Quebec (or Wisconsin) inbred 703. The standard inbred of this series has a white cob and strong red pericarp colour on the sides of the kernels with a colourless crown. A small F_2 from Pa703Y x Q703 has segregated 18 coloured pericarp-red cob, 10 coloured pericarp-white cob and 8 colourless pericarp-red cob. Both coloured vs. colourless pericarp and red vs. white cob show a good fit to a 3:1 ratio and the data would seem to indicate that the pericarp colour of Q703 or the red cob colour of Pa703Y is not the result of an allele at the pericarp-cob colour locus on chromosome 1. The absence of any colourless pericarp, white cob segregates is likely the consequence of the small population, since only two would be expected on the hypothesis of non-allelism.

It is difficult to envision the development of Pa703Y by simple mutation from Q703 since both cob and pericarp colour have changed (the cob colour presumably from recessive to dominant) and these appear to be dependent upon independent loci.

Robert I. Brawn

4. Mutational behavior of dark variegated pericarp.

It has previously been reported (N.L. 33, p. 73, 1959) that more red ears occur in the progeny of dark variegated, a new phenotype in the mutational spectrum of the P^{VV} allele, than in the progeny of the parental medium variegated. In 1960 a selfed ear which could appropriately be classified as very dark variegated or even very-very dark, was found in a family segregating dark variegated in inbred W9 background. There were no spots of red on this ear exceeding one kernel in size. The progeny of this ear is as follows:

coloured pericarp-red cob				coloured pericarp-white cob	
self red	very dk. var.	dark var.	medium var.	lt. var. or homo.	total
65 (35%)	104 (55%)	10 (5%)	5 (3%)	4 (2%)	188

The high proportion of red in the progeny of this phenotypically very dark variegated ear is consistent with the hypothesis of Brink and his students that the amount of red striping of the pericarp is related to the frequency of self-coloured offspring. However, the shortage of a corresponding lighter variegated class of progeny is difficult to explain.

This ear was apparently homozygous for pericarp colour since there were no colourless pericarp segregates. The red cob colour of the parental ear and of most of its offspring is inconsistent with the accepted hypothesis of pericarp-cob colour. Furthermore, the high proportion of self-coloured reds in the progeny is even more striking when one remembers that it has been established that the rate of change of medium variegated to red is lower in the progeny of homozygotes than in the progeny of heterozygotes.

Robert I. Brawn

5. Modulator activity of dark variegated.

Plants with dark variegated and medium variegated pericarp were used as males on a C Ds-tester. It would appear that the "mutations" at Ds are earlier and fewer in number (i. e., a coarser pattern of coloured and colourless areas is observed) when a Modulator from dark variegated activates Ds, than when a Mp from medium variegated is used. This would suggest that the difference between dark and medium variegated pericarp is a function of Mp. McClintock and Brink have both reported that increasing the dose of Ac or Mp delays and/or partially inhibits the changes at Ds and P resulting in a finer grade of mottling or more widely spaced red stripes. On this model of Ac-Mp action the dark variegated phenotype would result from a change of state of Mp in the direction of a lower dosage than that of the standard Mp of medium variegated.

This is at most a tentative hypothesis for the number of ears involved is small (5 and 4 respectively) and the C Ds-testers were different, although related, for the two categories of crosses.

Robert I. Brawn

MARQUETTE UNIVERSITY
Milwaukee, Wisconsin

1. The etched phenotype in the endosperm.

The etched allele (et) discovered by Dr. L. J. Stadler in an irradiated progeny has recently been examined for phenotypic detail in sectioned endosperm of et/et individuals. The following observations have been made:

1. The irregularly placed and irregularly shaped depressed areas on the surface of the kernel are not due to death of cells at these sites as one might presume from superficial observation.

2. Both the pericarp and aleurone layers at the depressed sites appear not to differ from Et/Et material. The pericarp is separated from the aleurone at these sites on the kernel leaving an air space which conceals the aleurone below (the detection of the etched spot is thus facilitated in a stock of colored aleurone).

3. The cells in the endosperm proper, underlying these depressed areas, are of a distinctly different type than the cells in the surrounding areas. They differ in that they are completely void of starch grains whereas the surrounding cells are normally packed with starch.

4. These starchless cells occur as well defined sectors in the endosperm, i. e., narrower toward the center of the kernel and broader toward the periphery.

5. The depression at the surface of the kernel results then from the starchless cell sectors occupying less space in the mature kernel than the adjacent areas filled with starch.

A possible interpretation of these findings is that in the et/et genotype the leucoplasts do not divide at a high enough rate to keep pace with cell division. Segregation of the leucoplast during cell division would then result in some cells being void. A cell, once void of the plastid would then be expected to give rise, by continued division, to a lineage of cells which lack leucoplasts. Repopulation of the leucoplasts could then take place in cells with at least one remaining when cell division ceases.

An alternative explanation is that the starch synthesis of the leucoplast is impaired though their division rate is normal. Spontaneous changes (mutations?) in the leucoplast could account for finding only some cell lineages exhibiting the starch storage defect. At the resolving power of these observations the presence of leucoplasts would go undetected.

It is of importance to note that et/et individuals also exhibit a virescence in the seedling (as reported by Stadler). A common basis for the chloroplast defect and the lack of starch in cells of the endosperm (leucoplast defect) is highly probable. The stage of development of the sporophyte and/or the physiology of the cells in which the etched phenotype occurs may account for the differences in response of a single cytoplasmic organelle to the genome. The granules of pigment (chromophores) in the aleurone show no alteration in the kernels examined.

These observations provide an explanation for the zygotic semi-lethality of et/et genotype noted by Dr. M. M. Rhoades (MNL 35, p. 67). If, during the development of the endosperm, the leucoplast is lost or becomes defective early enough there would remain an insufficient supply of stored food material for the embryo.

Irwin M. Greenblatt

Note: The author wishes to express his gratitude to Dr. S. Chase of the DeKalb Agr. Assoc., DeKalb, Ill., for providing both winter and summer research field space so that a maize genetics endeavor may be continued at an urban university.

UNIVERSITY OF MINNESOTA
St. Paul, Minnesota
Department of Agronomy and Plant Genetics

1. All arms tester interchange set in A188 inbred.

The following interchanges in the set have been isolated in homozygous condition: 1-3 (5883), 1-3 (5982), 1-9b, 2-4b, 2-4L, 2-6b, 2-6d, 3-4 (5156), 3-7c, 5-7e, 5-8a, 5-10 (5290), and 5-10 (6061). These have been crossed to the interchange set for identifying chromosomes as a check at the end of the backcrossing program. As more of those in the set reach the desired number of backcrosses, homozygous lines for them will be established and checked. This set is the one which we started introducing into A188, and subsequent backcrossing was continued by Dr. Jenkins and Dr. Sprague. At least two of the lines have been somewhat more difficult to use because they have only about 25% sterility. Other interchanges have been substituted for them.

C. R. Burnham

2. Dp-Df transmission tests for In 2c.

A stock homozygous for chocolate pericarp (Ch) and the paracentric In 2c has been established. Tests were made for possible transmission of the Dp-Df in plants heterozygous In 2c homozygous chocolate by crossing them as ♀ with ch ch. The 1442 progeny were all chocolate, none with colorless pericarp expected from functioning of Dp-Df (the Ch locus is distal to the inversion).

C. R. Burnham

3. A test to recognize Dp+Dp combinations from interchange crosses of type 2b (Gopinath & B. Genetics 1956).

Since duplications may be of use in modifying chemical composition associated with endosperm or other characters, it is desirable to have methods for identifying individuals carrying the duplication. One method is the following: 1. Cross the two interchanges that are homozygous for the Dominant allele at the locus to be duplicated. 2. The F₁ between them is crossed to a stock of either parent interchange which is homozygous for the recessive allele. 3. In the progeny any plants suspected of carrying the Dp+Dp may be tested by crossing them as ♀ to the double recessive. Plants carrying the duplication should give a ratio of about 3 dominant:1 recessive, and should have about 25% spore abortion. I am not aware that this test has been proposed, but I would be surprised if it hasn't. One feature of establishing a duplication by this method of using interchanges is that the duplicated region is not in tandem, but is in a different chromosome. One possible difficulty in getting it homozygous is that the duplication may show low transmission through the pollen.

C. R. Burnham

4. Inversions.

The inversion stocks isolated by Anderson and Longley were grown and crossed with W23. Forty-two of those listed in their Table 7 (ARS 34-16) were in the collection received from them.

5. Miscellaneous stocks available.

1. (Ra Ra) gl₁ v₅
2. Multiple recessive bm pr ys virescent with expanded glumes.

6. Progress in producing multiple interchange stocks.

Stocks homozygous for the following interchange combinations were produced: 2-1-7, 1-2-6, 1-3-7, 1-3-9, 3-2-6, 4-2-6, 4-2-8, 3-4-8, 4-6-5, 3-6-5, 6-5-7, and 8-10-9. Crosses were made with the chromosome identification set to check on the interchanges present in these and in the lines produced earlier. A stock homozygous for the 3-2-4-9 interchange combination has been established. The cross with 9-10b produced a 010. This F₁ has been backcrossed to 3-2-4-9 to add chromosome 10. A stock of 3-2-4-9-10 when crossed with 1-5-6-7-8 (already established) should produce plants with 2010. From this we expect to establish a stock homozygous for 3-2-4-9-10 plus 1-5-6-7-8. This is to be X-rayed in an attempt to unite the two rings.

Progress continues on the Inman plan which uses crosses between lines which have an increasing number of interchanges in common.

C. R. Burnham and Paul Yagyū

7. Segregation for quantitative characters in crosses with multiple interchange stocks.

Tests for possible association between quantitative characters and a 06 and a 08 were repeated in 1961. The general plan was to test F_1 's made up as (Inbred A x 06 Inbred B) and also as (Inbred B x 06 Inbred A). Parents, F_1 's, F_2 's and backcrosses to each parent were grown in a trial with 4 replications. Growth conditions were much more favorable than in 1960 (Newsletter #35 p. 87).

A preliminary examination of the data shows a difference for one of the F_1 's but not for the other for height to base of tassel. This showed a significant difference in the 1960 trials.

Paul Yagyū and C. R. Burnham

Assisting in the above work also were Ken Kasha, Jerome Arnold, and Gerald M. Welch. The work with multiple interchanges and related studies was supported by a Rockefeller Foundation Grant.

8. Dominance of genes controlling grain yield in corn.

Comstock and Robinson (1952) outlined an experimental approach for investigating level of dominance in the action of genes controlling quantitative traits which utilizes populations derived from crossing two homozygous lines. They pointed out that linkage equilibrium of genotypic frequencies cannot be anticipated in early generations of such a population. They further demonstrated that estimates of genetic variances would be affected by linkage disequilibrium so that the proposed measure of dominance would be biased upward until equilibrium was established. In order to investigate the effect of linkage disequilibrium upon estimates of dominance they recommended that data be obtained for the same single cross population in the F_2 generation and again in later generations when linkage equilibrium will have been approached.

This approach has been effectively employed at North Carolina (H. F. Robinson and co-workers) and at Nebraska (C. O. Gardner and J. H. Lonnquist) in studies on grain yield. Overdominance would have been inferred on the basis of F_2 generation data in these studies if the possible effect of linkage had not been considered. However, results from their advanced generation evaluations conclusively indicate that linkage disequilibrium existed and had the anticipated effect. Level of dominance estimates in the most advanced generations studied were fully compatible with the hypothesis of only partial dominance at all loci. However, these results do not preclude the possible existence of a range of dominance effects, i.e., partial dominance at many loci, but with overdominance at a sufficient number of loci to be of consequence with respect to population dynamics. The purpose of this study is to obtain more decisive information to distinguish between these two possible situations.

The experimental plan is to augment the approach reviewed above by selection in such a way to shift gene frequencies so that loci exhibiting partial to complete dominance will contribute progressively less to the results. Initial gene frequencies at segregating loci will be 0.5 in populations derived from crossing two homozygous lines. Continued selection should cause frequencies for the non-overdominant loci to approach either 1.0 or zero. In the case of overdominance selection favors the heterozygote so that gene frequency approaches an equilibrium value that will be in the range 0.2 to 0.8, unless the heterozygote advantage is very slight. If overdominance (of genes affecting grain yield) is present in more than a trivial amount its detection in this way will be more probable. Conversely, if negative results are obtained, the case against overdominance will be enhanced.

F_2 generation backcross matings have been made in two single cross populations for evaluation of genetic variances and corresponding level of dominance for grain yield in 1962. The population exhibiting the greatest level of dominance will be continued for this study. Following advancement to the F_3 generation this population will be divided into two groups: a control group which will be advanced by sib mating and a select group which will be subjected to full sib progeny test in every other generation of sib mating. Effectiveness of selection for increased grain yield will be determined in field trials following each cycle. The F_2 and advanced generations of the control group will be evaluated for estimation of level of dominance. The control and select groups will be evaluated for comparison of dominance estimates upon completion of three selection cycles when both groups are in the F_{11} generation. Completion of this study is expected to require a minimum of five years with the utilization of an overwinter nursery.

J. C. Sentz

9. Inheritance and linkage relations of genes for a serpentine character in *Zea mays*

An S_2 culture from the cross A495 x Red 30 made in 1958 produced plants with varying degrees of undulation in the lower portion of the stem. The extreme type has a serpentine appearance. The A188 interchange series is being used to determine location of the gene(s) controlling this character.

Alejandro Violic
E. H. Rinke

10. Association test between interchanges and multiple ear character.

Inbred E41 produces two almost identical ears per stalk. The pedigree indicates the multiple ear character was derived from Minnesota 13 variety. In order to determine the locus (loci) responsible for this character the 22 stocks of the all arms interchange tester series in A188 background (selected by Burnham and Longley) were crossed with E41 with the heterozygous interchange stocks as female parents. Semisterile F_1 's were backcrossed to A188 and also crossed to F_1 's within the stock.

John K. Lim
E. H. Rinke

11. Inheritance of reaction to Diplodia and Gibberella stalk-rot fungi.

Two single crosses involving susceptible by resistant inbreds, A239 x E11 and A427 x E10, are being used in this study. The parental inbreds, F_1 , F_2 and first backcross generations were infected by the toothpick method and their reaction recorded after splitting the stalks. Powers' partitioning analysis will be used to analyze the data.

R. E. Anderson
E. H. Rinke

12. Components of genetic variance for ear number.

Information concerning genetic variability and inheritance of traits that may contribute to grain yield is important in selecting for maximum production. The within population variance for ear number is being estimated for the five inbred lines, B14, Oh43, W22, Mt42 and A547, and the F_1 , F_2 , first backcross and second backcross generations derived from single crosses between these lines. Estimates of additive and dominance genetic variance will be made following general procedures first given by Mather (Biometrical Genetics, Dover Publications, 1949).

Charles Laible
J. C. Sentz

MISSISSIPPI STATE UNIVERSITY
State College, Mississippi
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1. A possible new knotted leaf similar to the original was found in an advanced generation from an F_2 ear of Dixie 18 double cross. It differs from knotted leaf only in the time and manner of expression. The character is not expressed on the leaves below the first visible ear shoot nor on the terminal leaf, and only from the ligule to approximately the mid-point of the affected leaves. Studies of the character are in progress.

C. O. Grogan
Patricia Sarvella

2. A new ear mutant in the form of an appendage ear was attached to the end of the main ear. The appendage was completely inverted to form a structure similar to a geode with seeds on the inside. The silks grew out through the end of the structure where they could be pollinated. In the following generation the inverted appendage ear was again observed, and some of the main ears showed signs of starting to invert near the apex.

C. O. Grogan
Patricia Sarvella

UNIVERSITY OF MISSOURI
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Department of Field Crops

1. Chemical treatment of seeds heterozygous for Wd and Yg₂.

In this experiment Yg₂ yg₂ and Wd wd heterozygous seeds were soaked in the dilute chemicals which had been found or are suspected to be mutagenic in lower and in higher organisms, frequently stirred, and rinsed after 8 hours of treatment at 23° C. The loss of the dominant genes or the absence of their function was observed phenotypically as yellow green or albino sectors on green background. The Wd wd heterozygote was produced by crossing a wd wd male containing Yg Wd C^I as an independent ring (obtained from Dr. G. Y. Kikudome) on a stock having normal chromosome 9 with the genes Yg Wd C. The role of the ring is to maintain the wd wd homozygous deficient stock. The ring is frequently lost during division resulting in albino stripes and a high proportion of gametes that are wd. For treatment the fully colored seeds were used since they probably lacked the ring. However it should be noted that rings which may have lost a segment of the ring carrying C^I during division, but retained either Wd, Yg or both are not detectible by this technique.

The following can be concluded from the table:

1. Ethyl methanesulfonate (EMS) and Diethyl Sulfate produce sectors in both leaf and shoot, the frequency being highest in the first leaf and linearly decreasing in subsequent leaves. The sectors are the smallest on the first leaves gradually increasing in size on subsequent leaves.

2. The mutagenic effect of Glycidol, Epichlorohydrin, Colchicine, although approaching significance, remains uncertain.

3. The frequency of sectors is of the same magnitude in both heterozygotes. This observation supports the assumption that colored seeds were Wd wd heterozygotes since, had the functional rings devoid of C^I carrying the genes Wd, Yg or both been frequently transmitted, the heterozygote seedlings could not be uniformly sectored.

4. In the Wd wd heterozygote one would expect albino sectors to be formed in excess since a break between the Wd locus and the centromere should give albino sectors and should be more frequent than breaks between Wd and Yg. On the contrary the Wd wd heterozygotes produced an excess of yellow green sectors. This observation indicates that EMS either preferentially breaks the chromosome toward the end, or produces a high proportion of mutations as opposed to chromosome breaks. Present data do not permit proper distinction between these possibilities.

Mutagenic effect of chemicals at the Yg and Wd loci

Chemical	Conc. M/lit	Lo-cus	No. of seeds treated	Surviving seedlings (%)	Av. Height 27 days after treatment (cm)	% of leaves sectored; frequency index ¹ of sectors per leaf (f) and size ² (s)						
						1st leaf % fs	2nd leaf % fs	3d leaf % fs	4th leaf % fs	5th leaf % fs	6th leaf % fs	7th leaf % fs
Ethyl Methanesulfonate	0.05	Yg	101	100.0	40.4	100 7a	100 6a	100 5a	93 4ab	** 67 3bc	**56 3bcd	*32 2bc
	0.05	Wd	30	100.0	39.2	100 6a	100 5a	100 5a	90 4ab	** 93 3bc	**29 2bc	*33 2bc
	0.10	Yg	50	92.0	35.0	100 7a	100 6a	100 5a	100 4ab	** 96 3bc	**73 3abc	*71 2bcd
	0.10	Wd	30	96.6	31.5	100 7a	100 6a	100 5a	100 4ab	**100 3bc	**67 3ac	*50 2bcd
	0.20	Yg	20	5.0	8.0	100 7a	100 6a	100 5a	100 4b	100 3bc	-- --	-- --
Diethyl Sulfate	0.05	Yg	100	19.0	13.2	74 5a	74 5a	82 4a	94 4ab	90 3ab	33 2ab	-- --
	0.10	Yg	49	0.0	--	-- --	-- --	-- --	-- --	-- --	-- --	-- --
Glycidol	0.05	Yg	100	92.0	38.0	17 1c	0 --	0 --	0 --	0 --	0 --	0 --
	0.10	Yg	42	89.7	33.9	100 1c	0 --	0 --	0 --	0 --	0 --	0 --
Epichlorohydrin	0.05	Yg	100	69.0	34.9	5 1c	0 --	0 --	3 1a	* 10 1a	*10 1b	*10 1b
	0.05	Wd	30	100.0	36.4	7 1c	0 --	0 --	*0 --	* 0 --	* 0 --	* 0 --
	0.10	Yg	49	59.2	31.2	4 1c	0 --	0 --	0 --	* 0 --	* 0 --	* 0 --
	0.15	Yg	30	0.0	--	-- --	-- --	-- --	-- --	-- --	-- --	-- --
	0.50	Yg	20	20.0	28.0	0 --	0 --	0 --	0 --	25 1b	25 1c	0 --
Colchicine	0.01	Yg	100	92.0	42.8	10 5a	10 4a	10 4a	8 3ab	1 1b	1 1b	0 --
	0.01	Wd	47	100.0	36.1	7 5a	7 4a	7 4a	7 3ab	5 1b	2 1b	0 --
	0.02	Yg	50	92.0	39.9	13 5a	13 5a	13 5a	9 4ab	2 1a	0 --	0 --
	0.02	Wd	49	91.8	34.8	11 5a	11 5a	11 4a	7 4ab	2 1b	0 --	0 --
Control	--	Yg	200	98.4	40.5	0 --	0 --	0 --	0 --	* 0 --	* 0 --	* 0 --
	--	Wd	70	100.0	38.6	0 --	1.4 1b	0 --	0 --	* 10 1a	* 0 --	* 0 --

(See next page for footnotes.)

¹Frequency index: (7) = 101<, (6) = 51-100, (5) = 11-50, (4) = 4-10, (3) = 1-3, (2) = 1-2, (1) = 1 sector per leaf respectively.

²Sector size (a) -- 1 mm wide, length from 1 mm to 3/4 length of leaf; (b) -- <1.5 mm; (c) -- 1.5-4 mm; (d) -- 4-10 mm wide, and extending from base to tip or to margin of leaf.

*Random sample (10-20% of surviving seedlings).

**Random sample (50% of surviving seedlings).

G. Ficsor

2. Transposition of mutability between components of the A_1 locus.

In studies of two separate cases of mutability arising from potentially compound alleles of the A_1 locus mutant types have occurred which suggest transposition of the factor responsible for mutability from one component to the other.

The first of these (MNL 30:101) originated from A^b and appeared to be composed of a stable α and a mutable recessive β component (β^m). The instability is controlled by one or more separate and as yet unidentified factors. The $\alpha \beta^m$ complex usually behaves just as would be expected on the basis of its structure. The α component may be removed by crossing over to produce a colorless mutable allele β^m , the β^m may change to recessive stable to produce a pale stable allele, the β^m may change to dominant stable and thus produce a reconstituted A^b ($\alpha \beta$) or the β component may change its state of mutability to produce a more or less unstable allele.

An occasional exception is found in the occurrence of cases where the mutability appears to have transferred to the α component leaving the β component in a recessive and much less mutable condition. Seeds of such a type are colorless with many pale and a few full colored sectors.

The second case originated several years ago in L. J. Stadler's cultures from the standard a^p allele. The new allele designated a^{pm} arose from a less mutable allele designated a^{px} which in turn arose from a^p . Because of its phenotypic expression and its failure to respond to attempts to subdivide it by crossing over the a^p allele was considered to be a single unit allele similar to the α' component of A^b Peru. However when one considers the behavior of its descendant allele a^{pm} one is led to conclude otherwise.

Regularly and without the need of any known mutator factor a^{pm} which has pale aleurone, red brown plant and dominant brown pericarp color (α phenotype) changes to A^r which has a purple seed, purple plant and recessive red pericarp color (β phenotype) or to A^{br} which has purple seed, purple plant and recessive brown pericarp color. Thus the mutants produced fail to fit into a linear series expected if a one-component locus were involved nor do they fit a two-component locus since the mutants obtained require that both components change in opposite directions at the same time. This seems quite illogical until one considers the possibility that such a dual change can occur if one component gives up something at the same time that the other gains

something or in other words that \underline{a}^{pm} is $\underline{\alpha} \underline{\beta}$ (incomplete) and that when \underline{a}^{pm} mutates to \underline{A}^r some element leaves $\underline{\alpha}$ and moves to the incomplete $\underline{\beta}$ component providing a complete $\underline{\beta}$; thus \underline{A}^r is $\underline{\alpha}$ (incomplete) $\underline{\beta}$ and is potentially able to revert to \underline{a}^{pm} again.

Regardless of the kind of interpretation it is clear that the controlling element of the \underline{a}^{pm} allele is able to move frequently from one aspect of \underline{A}_1 expression to another and that sometimes it affects both at the same time as shown by the presence of seeds that are simultaneously mutating from colorless to pale and from colorless to full color.

M. G. Nuffer

3. Location and effects of \underline{c}_2 .

The following data show \underline{c}_2 to be on chromosome 4:

Parent	X Y	Phase	++	+y	x+	xy	Recomb.
wx T4-9g/ \underline{c}_2	\underline{C}_2 -Wx	RS	117	30	46	4	35 ± 6.1%
su/ \underline{c}_2	\underline{C}_2 -Su	RS	617	205	251	63	46 ± 2.3%

If \underline{c}_2 is in the long arm it is probably beyond \underline{gl}_3 ($\underline{su-g}_1$ is around 35 units). The short arm has not been eliminated, however.

A few effects of \underline{c}_2 were described briefly in News Letter 34:91. A more complete summary is now possible. The homozygous recessive $\underline{c}_2 \underline{c}_2$ and double-mutant combinations with most others (\underline{a}_1 , \underline{a}_2 , \underline{bz}_1 , \underline{bz}_2 , \underline{c}_1 , \underline{C}^1 , \underline{r} , and \underline{pr}) have completely colorless aleurone tissue but \underline{c}_2 in kernels have dilute purple color. In plant tissues, \underline{c}_2 results in much-reduced pigmentation in the husks and sheaths; strong color develops only in the leaf auricles, glume bars, and similar tissues. The combinations of \underline{c}_2 with other plant-color factors show the effects of both; for example, $\underline{c}_2 \underline{a}_1$ plants (with $\underline{B} \underline{P}_1$) are very weak brown, like \underline{a}_1 plants in color but like \underline{c}_2 in strength of pigmentation. If \underline{c}_2 affects pericarp color in \underline{P} background at all, it is only by a very slight reduction in color intensity. The dosage effect of \underline{c}_2 in the aleurone is very clear; from a selfed ear of $\underline{+}/\underline{c}_2$, 13 selfs of full-color seeds were found to include 10 $\underline{+}/\underline{+}$ and 3 $\underline{+}/\underline{c}_2$, while 15 selfs of pale seeds were all $\underline{+}/\underline{c}_2$.

E. H. Coe

4. Resistance of B' to selection.

Selection for plant color for three generations has not altered the conversion-type pattern of inheritance followed by B'. A pl series using a uniform B pl parent (color grade 5 to 6) as the common recurring parent and a Pl series using a uniform B Pl parent (grade 7 to 8) were developed as light and dark lineages through three generations of selection. Selections were made in progenies of 30 or more individuals at each stage for each lineage and series. These lineages were planted in a randomized coded pattern and were graded for plant color without knowledge of pedigree. Cob color also was graded in the Pl series.

Lineage	Grade						Average grade
	0	1	2	3	4	5	
pl light, plants	26	84					0.76
pl dark, plants	15	110					0.88
Pl light, plants			6	78	27		3.19
Pl dark, plants				52	55	10	3.64
Pl light, cobs		30	77	4			1.77
Pl dark, cobs		23	87	5			1.84

E. H. Coe

5. Classification for B expression.

The gross tissues, husks, sheaths, and tassels, are useful in virtually any background to distinguish B from lower alleles but not to distinguish the lower alleles from each other. The bar of color at the base of the glumes in the tassel will permit distinction between b, which shows no bar, and higher alleles, all of which show at least bar color (excluding the mutable, B^v, which is phenotypically b except in sectors with full B expression). The cob, however, has several advantages over other tissues. It is protected from direct environmental influences (including bleaching and leaching), is the structure normally harvested and stored, and is not excessively fragile or bulky. The only major requirement for cob color expression is the purple (Pl) factor. Cob color grades give consistently better results than other gross tissues in predicting progeny phenotypes where distinction between B and B' and between different levels of B' is desired. Alleles such as B^b, which effect pigment synthesis in the tassel glume bar and parts of the culm, also elicit weak pigmentation in the hard, smooth parts of the cob, so they are distinguishable from b here as well as elsewhere.

The most convenient expression of B and its alleles, however, is in the coleoptile. Seedlings of B Pl constitution develop intense coleoptile color but so do R^r or r^r seedlings in the absence of B, so the genetic background for B classification should be R^g Pl or r^g Pl. In heated sandbenches, B color is expressed fully within two weeks from planting if sunlight has been adequate during the last few days. The seedling expression of B^b and B' is largely restricted to occasional tiny streaks on the coleoptile and these may be absent on some seedlings. Seedlings of b constitution are completely green in the R^g Pl background.

E. H. Coe

6. B' effect in the presence of a heterozygous translocation.

Plants from B¹/B T2-9a x B are indistinguishable from B¹ plants derived from crosses without the translocation. T2-9a has breaks at 2S.36 and 9L.58, with the map location in chromosome 2 to the right of sk, according to one small test by Patterson (Newsletter 26:10).

E. H. Coe

7. Bronze mutants and their action in anthocyanin synthesis.

The gene action sequence of A₁, A₂, C₁, R, In and Bz₁ in anthocyanin synthesis has been constructed, using complementary interactions between aleurone tissues (Newsletter 35:95). The position of action of bronze genes based on these observations was inconclusive. The bronze testers have anthocyanin pigment and when they are combined with colorless mutants, the colorless mutants develop pigment, leaving the possibility of simple diffusion of pigment rather than the required substrate transfer in anthocyanin synthesis. To eliminate this possibility of pigment diffusion, some further experiments were performed.

The double recessives r bz₁ and r bz₂ paired in the four possible combinations with singly recessive bz₁ and bz₂ were subjected to the previously described standard conditions. When colorless r bz₁ and r bz₂ are combined with bz₁ or bz₂ testers, only r bz₁ in the r bz₁:bz₂ pair develops pigment, while the others remain colorless, indicating that Bz₁ precedes Bz₂ in sequential action in anthocyanin formation and that the action of Bz₂ follows R and Bz₁ in anthocyanin synthesis. Further, double recessive bz₁ bz₂, although colorless, causes pigment to develop in a₂ and other testers, indicating that the bronze factors act after the others. These observations clearly show that the simple diffusion of anthocyanin pigment is not involved, at least from bronze mutants. Finally this method, i.e., use of double recessives, allows placement of the modifier genes in the action sequence in anthocyanin synthesis.

The action of C₂ in the sequence has not been established definitely, as the c₂-mutant in some cultures gives pigment by itself. All these observations, including the previous findings, establish the following sequence:

C^I, C₁, R, (In), A₁, A₂, Bz₁, Bz₂ ----- anthocyanin

Some of the preliminary studies of extraction of substrates with various solvents, acetone, alcohol, acid-alcohol etc. toward the direct demonstration of gene-enzyme relationship are encouraging but not convincing. It is possible that the characterization of these substrates can be expected to reveal the intermediates and the reaction steps in the biosynthesis of anthocyanin and may lead to further analysis of the mechanism of gene action and interaction in this system.

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UNITED STATES DEPARTMENT OF AGRICULTURE

1. Preferential pairing in trisome 3 plants with two standard chromosomes 3 and a chromosome 3 from exotic strains of maize.

Trisome 3 plants with the constitution of $A_1/a_1/a_1$ were produced by crossing trisome 3 plants homozygous for a_1 (on what shall be called standard chromosomes 3) with exotic lines which were homozygous for A_1 .

At meiosis when a bivalent and a univalent are formed, the two standard chromosomes 3 will tend to form the bivalent, while the exotic chromosome 3 will tend to be the univalent, if preferential pairing occurs. The univalent chromosome may be lost or it may go to either pole, in which case it is included in a disomic spore. Disomic pollen functions only rarely in fertilization. Consequently, if there is a greater than random frequency of exotic chromosomes 3 as univalents, there will be a frequency of A phenotypes in the progeny of less than $1/3$, when the trisome is used as the pollen parent.

Table 1. Backcross data of trisome 3 plants used as the pollen parent.

Source of A_1 chromosome	No. of kernels			Ratio A : a	χ^2
	A	a	Total		
Standard	4580	9404	13984	1 : 2.05	
Gourdseed	1566	3279	4845	1 : 2.09	0.3
Papago flour	1240	2980	4220	1 : 2.40	14.6
Zapaluta chica	2142	5630	7772	1 : 2.63	63.9
Grande	336	898	1234	1 : 2.67	11.5
Reventador	1038	2454	3492	1 : 2.36	9.7
Avati tupi	1237	3805	5042	1 : 3.08	103.9

Assuming gametophyte factors are not responsible for the aberrant ratios, all the trisomes with an exotic chromosome exhibited preferential pairing to a statistically significant degree, except in the case of the Gourdseed trisome. The possibility that gametophyte factors on chromosome 3 may be influencing the data can be checked using the disomic sibs of the trisomes. The disomic sibs should give a 1:1 ratio if gametophyte factors are absent. This has been found to be the case in Gourdseed, Papago, and Zapaluta chica.

This preferential pairing is believed to be indicative of structural dissimilarity between standard and exotic chromosomes. The nature of these structural differences is unknown. They may be small inversions, interpolations of heterochromatin or teosinte chromosome segments, or perhaps many very small structural differences on the level of magnitude of a gene. Crosses of standard-exotic hybrids and the standard trisomes will be made in the greenhouse this winter. The progeny will be examined for the degree of preferential pairing. In this way it should be possible to determine something about the structural differences.

It is noteworthy that the Gourdseed trisome did not exhibit a significant degree of preferential pairing. The standard is closely related to corn belt maize. Gourdseed (or a variety related to it) is believed to be one of the progenitors of corn belt maize. The exotic strain which exhibited the most preferential pairing, Avati tupi, is from Paraguay and is probably the least related to the corn belt maize strains.

G. G. Doyle

2. The synthesis of an artificial allotetraploid corn strain:

An allotetraploid corn strain would breed true for chromosome number and thus aneuploidy which is responsible for much of the sterility in tetraploid lines could be eliminated. Also an allotetraploid would be a true breeding single cross hybrid and any genetic constitution which is favorable for tetraploid fertility could be stabilized.

There are three methods by which an allotetraploid strain of corn can be produced. A corn genome must be modified by chromosome structural changes so that it loses most of its pairing affinity with the standard corn genome. In previous issues of the News Letter (32, 33, and 34) the writer presented data on the reduction in pairing affinity resulting from one inversion, In 3a. It was found that in tetraploids with the constitution of In 3a/In 3a/N 3/N 3 structurally homologous chromosomes were paired 77% of the time. In trisomes heterozygous for In 3a the corresponding frequency was 75%. If pairing were at random these values would be only 33.3%. If a chromosome contained several inversions it is probable that the pairing affinity would be very greatly reduced. Recent work by Grell with triploid *Drosophila* indicates that this is true. The first method is therefore to produce chromosomes containing many inversions by crossing inversion stocks. The writer has acquired 65 different inversions from various sources and has made crosses of combinable inversions. Unfortunately most of the inversions are overlapping and therefore not combinable. Extensive irradiation work is being carried on in an attempt to increase the supply of inversions.

The second method approaches the problem of developing multi-inverted chromosomes by the irradiation of material which already has one inversion in the hope of inducing a second one on the same chromosome. Last summer pollen from In 3a/In 3a plants was given 1000 r and was placed on silks of trisome 3 plants with constitution of $\underline{a_1}/\underline{a_1}/\underline{a_1}$. If no new inversion was induced then the frequency of $\underline{A_1}$ (from the In 3a stock) in the backcross progeny of the trisome used as the pollen parent should be about 22%. If another inversion has been induced in chromosome 3 then this frequency should be less. The modified In 3a chromosome will be subjected to further irradiation to obtain a third inversion and so forth. Eventually a new chromosome 3 will be produced which will have very little pairing affinity for the standard chromosome 3. If this procedure works satisfactorily it will be done with the other chromosomes.

The third method is suggested by the results obtained with the exotic trisomes. While none of the exotic chromosomes exhibited enough differential pairing affinity to be used in an allotetraploid, it should be possible to find recombinant chromosomes in the progeny of hybrids of exotic strains which will exhibit more preferential pairing than either of the parents. For example a hybrid of Zapaluta chica and Papago flour corn will be crossed with the standard trisome 3. Recombinant chromosomes should show transgressive segregation for pairing affinity if these two strains have different structural rearrangements. Other hybrids will be used.

G. G. Doyle

3. Numerical non-disjunction in tetraploid corn.

Numerical non-disjunction is the 3 to 1 separation of four homologous chromosomes of a tetraploid at the first division of meiosis. This event results in aneuploidy in the offspring of a eutetraploid.

The frequency of numerical non-disjunction can be determined for a particular chromosome by crossing a quadriplex (AAAA) with a nulliplex (aaaa), and then progeny test the offspring. If numerical non-disjunction has not occurred the result is a plant with the constitution of AAaa which will give a testcross ratio of about 5:1. The products of numerical non-disjunction will be triplex and simplex (usually AAAaa and Aaa) which give testcross ratios of about 12:1 and 1:1, respectively. All these ratios can be easily distinguished. The frequency of numerical non-disjunction has been determined for two chromosomes, 2 and 9, using the Lg_1 and Wx loci, respectively. The initial crosses of quadriplex by nulliplex were made by D. L. Shaver.

Table 2. The frequency of numerical non-disjunction

Cross	Triplexes	Number of Duplexes	Simplexes	% Numerical Non-disjunction
$4n\ lg\ X\ 4n\ Lg$	2	83	1	3.5
$4n\ Lg\ X\ 4n\ lg$	5	239	9	5.5
$4n\ wx\ X\ 4n\ Wx$	4	332	1	1.5
$4n\ Wx\ X\ 4n\ wx$	2	93	5	7.0

G. G. Doyle

4. The duplication of specific chromosome segments by crossing translocations involving the same chromosomes.

The technique for the duplication of specific chromosome segments was first proposed by H. J. Muller (Journal of Genetics 23:299-334) in 1930. In 1956, Gopinath and Burnham worked out the problem in great detail (Genetics 41:382-395).

Pairs of translocations suitable for the duplication of chromosome segments containing the y, wx, ae, or su locus have been crossed with each other. It is hoped that the duplication of these loci will modify the chemical composition of the corn endosperm. Also some information about gene action should result from this work. If a recessive gene is an amorph, its duplication should have no effect. If a recessive gene is a hypomorph, then its duplication should result in a phenotype which approaches or exceeds the dominant phenotype.

G. G. Doyle

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1. A new gene to mark the distal end of the short arm of chromosome 6.

In a previous newsletter (No. 33 p. 102) a recessive ragged seedling character here designated rgd was shown to be on the opposite side of y from tsg and later to be to the left of y. F_2 repulsion data were obtained from 9425 seedlings from 29 ears with the genotype rgd y / + Y classified as follows:

4810 + Y : 2169 rg Y : 2409 + y : 37 rg y

which indicates approximately 13% recombination and places rgd very close to po.

In 1960, one selfed ear from a plant with the genotype + po Y / + /rgd + y tsg and 3 from plants with the genotype + po Y / rgd + y were obtained. Seeds were grown and plants which shed pollen were selfed. Plant classifications from these 4 F_2 progenies are in Table 1.

Table 1

Phenotype	60-4108-3	60-4108-5	60-4108-6	60-4108-10
+ Y +	88	70	126	54
po Y +	45	28	48	32
+ Y tsg	2	--	--	--
- Y -*	54	41	40	33
+ y +	0	17	41	15
po y +	0	1	1	3
+ y tsg	27	--	--	--
- y -*	43	31	36	27
Sum	259	188	292	164

* Seeds did not germinate or plants did not emerge.

Plants in the phenotypic classes Y and y were selfed. The numbers of plants selfed in each progeny and their genotypes with respect to Y y and rgd as determined by seedling tests are in Table 2. The genotypes are designated by classes 1 to 6.

Table 2

Class	Genotype	60-4108-3	60-4108-5	60-4108-6	60-4108-10
1	Y + / Y +	0	0	1	0
2	Y + / Y rgd	3	3	6	6
3	Y + / y +	8	3	8	3
4	Y + / y rgd	61	43	94	34
5	y + / y +	0	3	0	0
6	y + / y rgd	0	9	17	5
Total ears		72	61	126	48

The classes in Table 2 were established by germinating and classifying 20 seeds from Y Y and y y ears and 20 white seeds from each Y y ear. This procedure could result in placing Y rgd / y + ears in class 3. The data from white seeds of class 4 ears are in Table 3.

Table 3

1960 Ear	No. of 1961 progeny ears	+	rgd	Sum	% Recombinations*
4108-3	61	404	768	1172	19.1
-5	43	260	532	792	18.1
-6	94	602	1193	1795	18.5
-10	34	158	473	631	13.4
Sum	232	1424	2966	4390	17.8

* $p = 1 - \sqrt{\frac{d}{n}}$ where $d = y$ rgd class

The recombination values are consistent around 17.8% indicating rgd to be to the left of po. Classes 1 and 5 of Table 2, however, provide the critical comparisons.

With the genotype + po Y / rgd + y and assuming the order and distances to be rgd-4-po-11-y, a class 5 ear may be obtained by union of two gametes each with a single crossover, one of which must be in region 1. This will occur with an expected frequency of 4% of the selfed y y ears to give + po y / + + y or + + y / + + y genotypes. With the positions of rgd and po reversed, only .17% of selfed y y ears are expected to breed true for Rgd and this only if no chromosome interference is assumed. The observed proportion was 3/34 or almost 9%.

The single ear in class 1 is equally interesting. With rgd to the left of po a noncrossover and a double crossover are required as a minimum to give + + Y / + po Y. The expected frequency is 1% of the selfed Y Y ears. With po and rgd reversed a noncrossover gamete and a region 1 crossover gamete will give po + Y / + + Y and two gametes each

with a single crossover in region 1 will give $\frac{++Y}{++Y}$. The combined expectation of these two genotypes is 22% of selfed $\underline{Y Y}$ ears. The observed frequency is 1/19 or about 5%.

Thus both classes 1 and 5 in Table 2 as well as the recombination percentages in Table 3 agree in placing rgd distal to po on the short arm.

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1. Synthesis of hybrid esterase enzymes in E heterozygotes.

By investigating newly synthesized enzymes that are still associated with the template on which they were synthesized, we have been able to establish that in the heterozygotes the new hybrid enzymes are synthesized as such on the ribosomes and do not result from random dimerization of previously synthesized monomers. We have been able to rule out the possibility that the particle-bound enzymes represent nonspecific adsorption of free enzymes to the ribosomes. The hybrid enzymes very probably result from interaction between two messenger RNA molecules specified by the two alleles, each of which contributes some information to the specificity of the hybrid enzyme.

Drew Schwartz

2. Regulatory mutant at the E locus.

The esterase enzymes specified by the E alleles are normally synthesized in the maternal tissue, endosperm, and embryo of the developing kernel as well as in the young seedling. A mutant has been found which affects the distribution of the enzyme in these tissues. Normal amounts of enzyme are synthesized in the diploid tissue but synthesis of the enzyme in the endosperm is almost completely blocked. The mutant, designated $\underline{E}^{R'}$, controls the synthesis of an F type enzyme that shows the same electrophoretic mobility as the F type enzyme produced by the normally behaving \underline{E}^R allele. In heterozygotes the $\underline{E}^{R'}$ allele is not influenced by and does not affect the homologous allele so that, for example, in $\underline{E}^{R'}/\underline{E}^{R'}/\underline{E}^N$ endosperm only N type enzyme is detected electrophoretically and in the amount expected from a single dose of the gene.

Since the E gene is not active throughout the life cycle of the plant, we propose that the E locus is compounded of a regulatory and structural gene. According to this hypothesis, $\underline{E}^{R'}$ has a mutant regulatory gene which fails to "turn on" the structural gene in the endosperm tissue. The regulatory gene is similar to the operon in the β -galactosidase case described by Jacob and Monod in that it operates only in the cis condition controlling the structural gene on the same chromosome, and is very closely linked to the structural gene. No crossovers have been found in over 2000 tested endosperms. Since the E alleles can be distinguished only by the electrophoretic migration rate of the esterase enzymes which they specify, the test involves individual electrophoresis of single, immature endosperms.

Drew Schwartz

3. Two new alleles of the E gene found in teosinte.

Three alleles of the E gene, \underline{E}^F , \underline{E}^N , and \underline{E}^S , have been found in maize. In a study of 550 strains of maize from South and Central America, we found that the \underline{E}^N allele was the most common and the \underline{E}^F allele the least common. Two other alleles, \underline{E}^L and \underline{E}^R , have been found in some teosinte strains. \underline{E}^L and \underline{E}^R specify enzymes with electrophoretic migration rates intermediate between the enzymes specified by \underline{E}^F and \underline{E}^N , and \underline{E}^N and \underline{E}^S , respectively. The teosinte heterozygotes $\underline{E}^L/\underline{E}^R$ form a hybrid enzyme with an intermediate migration rate just as do the maize heterozygotes. A hybrid enzyme is also found in the \underline{E}^F maize \underline{E}^R teosinte hybrid plants. Other hybrid crosses are presently being made for further tests.

We have tested eight teosinte lines supplied by Dr. P. C. Mangelsdorf. Chapingo, Chilpancingo, Chalco, Arcelia, and Huixta carry only the alleles found in maize, \underline{E}^F , \underline{E}^N , or \underline{E}^S , while the Lake Retana, Florida, and El Valle teosintes carry only the new alleles \underline{E}^L or \underline{E}^R . Dr. Mangelsdorf has pointed out the interesting comparison that the latter three teosinte lines are quite Tripsacoid. Surprisingly, this esterase is not found in young *Tripsacum* seedlings. We have tested two species, *T. floridanum* and *T. dactyloides*. At the moment we cannot tell whether the absence of the enzyme is due to absence of the E gene or simply a different distribution of gene activity in the life cycle of *Tripsacum*.

Drew Schwartz

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1. Evidence for linkage of genes for stalk-rot susceptibility with genes located in chromosomes 7 of inbreds Mo21A and NC34.

Translocation studies (Agron. Jour. 49:197-201) have shown that major genes for resistance to *Helminthosporium turcicum* leaf blight are located in the short arms of chromosomes 7 of inbreds Mo21A and NC34. Tests designed to locate these genes more precisely were grown at Columbus, Ohio, using a chromosome 7 marker gene stock. Plants from the backcrosses ($o_2 \underline{v}_5 \underline{ra}_1 \underline{gl}_1$ x Mo21A) x $o_2 \underline{v}_5 \underline{ra}_1 \underline{gl}_1$ and ($o_2 \underline{v}_5 \underline{ra}_1 \underline{gl}_1$ x NC34) x $o_2 \underline{v}_5 \underline{ra}_1 \underline{gl}_1$ were inoculated with ground leaf inoculum of *H. turcicum*. Difficulties of classification made the gene \underline{v}_5 unusable.

Leaf blight failed to reach a high level of disease incidence by rating time. Although it appeared doubtful that good differences in disease reaction could be obtained, individual plants from each of the backcrosses were scored for *H. turcicum*. Statistical analyses of the data failed to show any significant differences between the various gene classes for *H. turcicum* reaction.

A natural infestation of stalk rot, most likely *Gibberella zeae*, caused many of the plants to die prematurely. These were noted as dead plants since it was difficult, as well as impracticable, to score them for leaf blight.

In working up the data, it became apparent that the greater proportion of dead plants occurred in the dominant parental classes. Frequency distributions for each of the gene classes of the two backcrosses are shown in Table 1. Chi-square tests for independence of dead and living plants in the parental classes yielded values with probabilities of occurrence well beyond the 0.50 per cent point. The data indicate that genes for stalk-rot susceptibility are linked with the dominant alleles of o_2 ra_1 gl_1 located in or near the short arms of chromosomes 7 of inbreds Mo21A and NC34.

Table 1. Frequency distributions by genetic classes of living and dead plants and the P values for chi-square tests of independence for the backcrosses (o_2 ra_1 gl_1 x Mo21A) x o_2 ra_1 gl_1 and (o_2 ra_1 gl_1 x NC34) x o_2 ra_1 gl_1 .

Genetic class	Mo21A Backcrosses			NC34 Backcrosses		
	Plants: living	Plants: dead	x^2 for independence	Plants: living	Plants: dead	x^2 for independence
	No.	No.	P value	No.	No.	P value
o_2 ra_1 gl_1 + + +	356 243	44 161	<0.005	339 209	75 185	<0.005
o_2 + + + ra_1 gl_1	39 34	10 11	≤ 0.50	25 45	16 9	<0.05
+ + gl_1 o_2 ra_1 +	4 4	4 0		5 4	10 1	
+ ra_1 + o_2 + gl_1	1 6	0 1		0 0	0 0	

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1. Parthenogenesis.

During 1959, 290 plants of the "Pa G-100 Synthetic" were bagged before silk emergence to prevent pollination in an attempt to follow up the report of S. H. Yarnell dealing with parthenogenesis in corn. The "Pa G-100 Synthetic" was constituted from numerous early to extremely early lines, mainly of Ottawa, Canada, and Wisconsin origin.

Seeds per ear developed under the bags varied from 0 to 239. A frequency distribution suggested random development of seeds that might have represented pollen contamination carried by insects or wind.

All seed was planted ear-to-row in 1960. Of the approximately 30 small 'inbred-appearing' plants, 26 were successfully selfed. The seeds from these 26 ears were planted in 1961 for a between plants within ears uniformity test. On the basis of segregation for cob color, kernel color, kernel flintiness, and kernel degree of dent 16 entries were eliminated. During 1961 several selfs of each entry were also made.

The 10 remaining stocks will be more carefully screened in 1962. This will mainly be based on variances within and variances between ears of entries. The lines W D, Co 106, Co 109, Co 110, and W 59E (important components of the original synthetic), the original synthetic, and four single crosses will be used for comparison.

G. W. Gorsline
Department of Agronomy

2. A computer method of double cross prediction.

A new program has been devised at the Pennsylvania State University Computation Center to predict the results of double cross hybrids. The program was written in FORTRAN and compiled on the IBM 7074 but is adaptable to any computer for which a FORTRAN compiler is available. The program can accommodate the single cross data of twenty inbred lines for eighteen or fewer variables. It features adjustable limits for each variable so that only prediction values above a chosen limit are included in the output. The table or card output includes a program title, experiment identification, designation of the inbreds and variable designation in addition to the prediction values. The computation time is too brief to estimate. This program is available on request.

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1. Segregation for a cyclic hydroxamate in maize seedlings.

Maize contains high concentrations of a phenol-like sweet substance (R. J. Suhadolnik, Ph.D. Thesis, Penn. State Univ. 1957; R. H. Hamilton, Ph.D. Thesis Mich. State Univ. 1960). The structure is now established to be 2,4-dihydroxy-3-keto-7-methoxybenzoxazine [A. I. Virtanen, P. Hietala, and E. Honkanen, Acta Chem. Scand. 114:502-507 (1960); R. H. Hamilton, R. S. Bandurski and W. R. Reusch, Cereal Chem. (in press)]. This cyclic hydroxamate may be a factor in resistance of maize to 2-chloro-s-triazine herbicides. [W. Roth and E. Knüßli, Experientia 17:312 (1961); R. H. Hamilton, and D. E. Moreland, Science 135:373 (1962)]. Also 6-methoxybenzoxazolinone (implicated in resistance to disease and corn borer) is a degradation product of this substance.

Seedlings of twenty inbred lines were examined qualitatively for the presence of the cyclic hydroxamate or its 2-glucoside; all were found to contain it. Selections by Dr. George Gorsline of an open pollinated synthetic, Gehr Yellow Dent, were inbred in 1959 and 1960 at Pennsylvania State University. One 1959 ear was found to be segregating (208 + to 59 -) for presence and absence (a trace present if pooled seedlings were extracted) of the cyclic hydroxamate. The open pollinated variety contained the cyclic hydroxamate (36+). One ear of the segregating 1959 population was selfed in 1960 and two ears were selected. One was all minus (33-) while the other ear was segregating (37+ to 8-).

R. H. Hamilton

2. Genetic iron-deficiency chlorosis in maize.

A. The yellow-stripe phenotype displayed by ys_3/ys_3 plants (MNL 35:111) is another iron-deficiency chlorosis. Plants of genotype ys_3/ys_3 have been grown in nutrient-solution, sand and soil cultures for a physiological comparison with ys_1/ys_1 plants similarly cultured. Seedlings of either genotype produced completely green leaves when sprayed with aqueous solutions of ferrous or ferric salts or when iron chelated as Fe-HEDTA (ferric chelate of N-hydroxyethylethylenediaminetriacetic acid) was incorporated into the rooting medium.

The metabolic lesion associated with the ys_1 locus appears to be localized to the absorbing areas of the roots. (Bot. Gaz. in press). When phosphate was deleted from the nutrient medium, ys_1/ys_1 plants produced fully green leaves. Chlorotic ys_1/ys_1 plants showed noticeable greening within 48 hours following an iron spray treatment, the addition of Fe-HEDTA to the culture medium, root tip removal in solution culture where iron was available to $+/ys_1$ but not to ys_1/ys_1 plants, or phosphate deletion.

In contrast, ys_3/ys_3 plants did not green rapidly when treated to correct the ys_1 -type chlorosis. A period of approximately six days elapsed following a foliar spray of aqueous $FeSO_4$ before correction of the ys_3 -type chlorosis was detected. When grown adjacently in a greenhouse soil bed of pH 5.5, ys_1/ys_1 plants remained yellow-striped whereas ys_3/ys_3 seedlings gradually greened. The metabolic lesion associated with the ys_3 locus appears to be more in the translocation or utilization of iron than in uptake. These alleles are being converted to a common background for a more definitive evaluation of these responses.

B. Approximately ten new selections of iron-deficiency chlorosis in maize have been observed and collected this past season; leaves of these yellow-striped plants responded by greening locally following spraying with an aqueous solution of $FeSO_4$ in the field or in the greenhouse. Of four selections grown in sand culture, all responded as did ys_1/ys_1 seedlings; Fe-HEDTA supplied in the applied nutrient solution induced full leaf greening. Seedlots yielding plants displaying this chlorosis were found in plant introductions grown by Dr. Roy Creech (P.I. 177591, 179561, 196127, 200296, 217461, and 231738), two inbred lines of Dr. C. C. Wernham and in an Italian flint inbred selected and supplied by Dr. Angelo Bianchi.

An acute yellow stripe appeared to be lethal in the field (3 plants perished with only scant greening). Two other chlorotic seedlings from the same row (16 green:5 chlorotic) greened completely after transfer from the field to sand culture with Fe-HEDTA in the greenhouse.

William D. Bell

3. Nutritional factors of maize mutants involving factors other than iron metabolism.

Tightly rolled leaves of adherent seedlings and plants have responded to foliar applications of a nutrient solution (Hoagland and Arnon's #1 minus iron). A 0.005 M CaCl_2 solution poured into the leaf rolls of such plants elicited an unrolling of the leaves but symptoms subsequently developed which appeared to be those of potassium deficiency. More leaf unrolling occurred when the application of the nutrient solution was accompanied by puncturing the main vascular bundles of the adherent leaves. A solution containing both 0.005 M CaCl_2 and 0.005 M K_2SO_4 was less effective than the nutrient solution in correcting adherence.

Albescent seedlings producing only white leaf tissue regreened in some cases following one or more transfers to aerated or unaerated complete nutrient solutions; sand-cultured seedlings of the same selfed seedlot continued to produce only white leaves. Greening of al seedlings seemed to be most pronounced when the attached grain was immersed in the nutrient solution. Applications of casein hydrolysate, yeast extract or coconut milk to mechanically exposed cotyledons of white al seedlings produced no beneficial effects. Leaf tip feeding with 0.3 M sucrose (method of Spoehr) prolonged the life of white al plants but induced no further visible chlorophyll formation.

A pale green selection segregating from selfed plants of P.I. 194047 has responded to foliar applications of Nu-Iron, a product of Tennessee Corp. Leaf areas which had been in contact with the spray became visibly greener in several days; untreated seedlings or those sprayed with solutions of FeSO_4 alone or in combination with micronutrients did not survive. Sprays of ferric oxalate solution did not produce the same effect nor were benefits observed following the incorporation of Fe-HEDTA or ferric oxalate in the solutions supplied to sand-grown pale green 194047 seedlings.

Comparable responses in unclassified pale green seedlings were observed in the field; when splashed with a clay loam mud either deliberately or during precipitation, a localized increase of the greening of leaf tissues resulted. Areas greened when in contact with the dried mud applied to either the adaxial or abaxial leaf surface. Iron and/or micronutrient solutions as sprays elicited no greening of these seedlings.

A selfed selection from P.I. 174415 was reported by Dr. H. H. Kramer (personal communication) to respond to a mixture of micronutrient solutions. The enhancement of greening of pale green, yellow, or white seedlings from this ear was confirmed using an aqueous foliar application of FeSO_4 with the micronutrients indicated by Hoagland and Arnon. A white seedling thus treated became green enough to produce selfed

seed; white and yellow seedlings appeared in the succeeding seed generation. Ferrrous sulfate or any of the micronutrients (B, Mn, Zn, Cu, Mo) alone as sprays did not produce results equivalent to the combination.

Unclassified chlorophyll-deficient mutants are requested for comparison with the above. No reports seem to be available on ys_2 which would be desirable to compare with the iron-deficiency mutants.

William D. Bell

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College, Laguna, Philippines
Department of Agronomy

1. Restorer factors in Philippine corn inbreds.

In an effort to eliminate detasseling in the production of F_1 double crosses, it was necessary to survey the ability of Philippine inbreds to restore pollen shedding in plants with Texas-type cytoplasm. Standard and promising flint inbreds extracted from College Yellow Flint and Cuban Yellow Flint and promising sweet corn inbreds extracted from Hawaii Yellow Sweet, Philippine Yellow Sweet, Maize Chiripo Dulce and Colombia Yellow Sweet were included in the test. Regardless of whether inbreds from individual varietal sources were treated separately or in bulk, equal frequencies of restored and unrestored plants were observed among the F_1 crosses of the flint corn lines to Texas-type cytoplasm sources. This strongly indicates that the two flint varieties used as inbred sources were originally heterozygous for the restorer factors.

Among the sweet corn inbreds only one line extracted from Philippine Yellow Sweet showed a partial capacity of pollen restoration (1:1 ratio of sterile to fertile plants in the F_1 and BC_1). The rest were non-restorers.

A. A. Gomez
A. C. Mercado, Jr.

2. The susceptibility of cytoplasmic male sterile lines of corn to *Helminthosporium maydis*.

Twelve standard Philippine inbred lines of corn which were sterilized through the incorporation of cytoplasmic male sterility derived from $F_{111}T$, an introduced inbred line from Florida, U.S.A., were compared to their respective normal counterparts for their reaction to *Helminthosporium maydis* at College, Laguna, in 1961 wet season. In all cases, the cyto-sterile inbred versions obtained after four to seven backcrossings were found to be much more susceptible to the disease than their normal inbred counterparts.

Male-cyto-sterile lines representing four single crosses and five double-crosses with one of these cyto-sterile inbreds as seed parent, consistently showed extreme susceptibility to the same disease whereas the normal counterpart manifested conditions of slight to moderate infection only.

A. C. Mercado, Jr.
R. M. Lantican

3. Development of sugary-waxy corn (su su wx wx).

The sugary-waxy inbreds reported in the 1961 issue were all either lost or discarded because they were extremely weak and very poor pollen shedders. This further strengthened the suspicion that unfavorable yield genes are linked to the waxy and sugary alleles and that the complementary effect of the two linkage groups results in very weak plants. In an effort to verify this, a new group of lines (already in the S₃) is now being developed from crosses of sweet and glutinous corn, and instead of maintaining the sugary-waxy plants, only the flint kernels from ears segregating for the three kernel types (flint, sweet, waxy) are selected for planting in the next generation. Four out of nine of these selected kernels are expected to be heterozygous for both loci. Thus, the genes controlling kernel types are always maintained in the heterozygous condition while the other factors follow normal inbreeding behavior.

After the general combining ability test four sublimes--flint, sweet, waxy and waxy sweet--will be extracted from each of the selected inbreds. These four will then be compared with each other for vigor and yielding ability. If all waxy-sweet lines shall consistently be inferior to the others, then it is highly possible that some unfavorable yield genes are indeed linked to the recessive alleles of these two loci.

A. A. Gomez
R. M. Payson
I. S. Santos

4. Susceptibility of glutinous corn to rust.

In our breeding nurseries and performance tests, where the different corn types are planted adjacent to each other, it has been consistently observed for the last five seasons that the glutinous type, as a group, is very susceptible to corn rust. In all plantings the glutinous group was always the first and the most heavily damaged by the disease. To verify this observation a separate and more systematic experiment on relative resistance to rust of the different corn types will be planted this season.

A. A. Gomez
R. M. Payson

5. Rust-resistant corn lines.

In an extensive screening for rust resistant corn plants at the Central Experiment Station, College, Laguna, 7 varieties introduced from Central and Northern South America showed a relatively high degree of resistance to the disease. Rust-resistant inbreds are presently being extracted from these varieties. Of the three inbreds (B38, Cuzco and GG 208) reported by Russell and Hooker of the U.S.D.A. to be resistant to corn rust incited by Puccinia sorghi Schw., only GG208 was observed to be highly resistant.

F. A. Aquilizan
A. A. Gomez

PIONEER HI-BRED CORN COMPANY
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1. Interactions between maize genotypes and teosinte cytoplasm.

The inbreds WF9 and C106 have been backcrossed 5 times, as males, into 10 different cytoplasmic sources. The 10 include 5 exotic open-pollinated varieties of maize, 4 inbred lines of maize, and a strain of "Florida" teosinte. One plant of each cytoplasmic source was used as source of cytoplasm for backcrossing to both WF9 and C106. At the fifth backcross all 9 of the strains with maize cytoplasm were identical in appearance to their pollinator parent. However, the C106 with teosinte cytoplasm and the WF9 with teosinte cytoplasm were markedly reduced in vigor throughout the growing season and produced plants with slenderer culms, narrower leaves, fewer internodes, fewer tassel branches and both strains were about a week late in flowering, compared to their pollinator parent. In general, the effect resembled that reported for teosinte cytoplasm by Mazoti (Rev. de Invest. Agric., 1954). Both strains also were partially female sterile; that is, no ears had more than a scattering of kernels, even when pollinated with plentiful supplies of fertile pollen. In addition, the WF9 in teosinte cytoplasm was completely pollen sterile. This may have been only an effect of reduced vigor on the naturally poor pollen shedding abilities of WF9. On the other hand, it may be that the teosinte cytoplasm used here has an interaction with certain genotypes which results in pollen sterility, independently of vigor effects. Appropriate crosses to test this hypothesis are being made.

D. N. Duvick

2. Rapid recurrent and reciprocal selection.

In Newsletter #33 p. 95 a modification of recurrent and reciprocal selection is described which utilizes simply inherited kernel characteristics displaying incomplete dominance to reduce the number of generations required to complete a cycle of selection. Since that time progress has been made on two studies associated with the scheme.

Seven white (A34, A177, A188, Ky27, NY2, 4Co.82, and 33-16) and seven yellow (A334, A375, NY3, Oh40B, Oh51A, Os420, and W25) inbred lines representing virtually 14 different open pollinated varieties were used in the studies.

A. The 98 possible crosses (including reciprocals) among the two groups were made and observed to examine the complexities of separating 3/3 yellow, 2/3 yellow-1/3 white, 2/3 white-1/3 yellow, 3/3 white endosperms necessary to segregate intercrossed from testcrossed seed via the previously described scheme.

Use of an Agron Color Control Instrument (courtesy of Kurth Malt-ing Co.) indicated perceptible differences in endosperm color between reciprocals of a given cross but was generally ineffective over the range of all the material studied. The writer's observation was that some 2/3 yellow-1/3 white endosperms were more yellow than other 3/3 yellow (viz. Oh51A crosses compared to Oh40B selfed, for example). All 2/3 white-1/3 yellow vs. 3/3 white differences appeared sufficient for separation. Evidently a recurrent selection program would be feasible

while a reciprocal selection program would present difficulties in separating kernel colors if a broad range of material were used.

As the scheme was previously outlined, the seed parent of the final hybrid would be used as a pollinator in the intercrossing and testcrossing block during selection; therefore, a desirable yellow seed parent could be the tester for a heterogeneous group of white endosperm material with no problems in seed color separation expected.

B. The 21 possible yellow x yellow, 21 possible white x white and 49 possible yellow x white crosses were grown in micro-tests to determine the relative merit of the three groups of germ plasm. The material was grown at a harvested stand of 13M plants per acre and averaged approximately 95 bu/acre. In terms of yellow x yellow equals 100%, white x white yielded 97%, and yellow x white yielded 103%. The differences among groups were statistically significant. Evidently a "built-in" increase in heterosis could be expected in a yellow x white program probably due to the genetic divergence between these groups.

A. Forrest Troyer

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and
TRANSVAAL REGION, DEPARTMENT OF AGRICULTURE
Pretoria, Republic of South Africa

1. Disease resistance of Mex 155.

The maize line Mex 155 (selected lines 86, 152 and 156), found to be highly resistant to *Helminthosporium* leaf blight at Pretoria in the Republic of South Africa, is reported to be highly resistant to leaf blight in France. Mex 155 is also highly resistant to downy mildew (*Sclerospora sorghi*) in the Republic of South Africa. It has a long growing season and good combining ability.

P. M. le Roux
Department of Plant Pathology

2. Seed treatment with organic mercury fungicides discontinued.

Organic mercury fungicides used as standard seed treatment are being replaced by Captan 75. Captan 75 is the only fungicide recommended for use on maize seed at present in the Republic of South Africa.

P. M. le Roux
Department of Plant Pathology

3. Are controlling elements episomic?

Controlling elements in maize are unique in that they can move spontaneously to a number of positions throughout the genome. The similarity of this behavior to that of episomic elements in bacteria has been pointed out by various authors. Episomic elements differ from controlling elements in that, in addition to occupying various chromosomal sites, they may also behave as cytoplasmic particles.

The author has been struck by the tendency of some families of light variegated stocks to become lighter and lighter due to an accumulation of Modulators. This is what might be expected if M_p was occasionally multiplying independently of the chromosomes. Since the variegated pericarp material has been maintained in heterozygous condition for many generations with a colorless inbred always used as the male parent, possible differences in reciprocal crosses would not have been apparent.

Various tests have been undertaken to search for cases of the cytoplasmic inheritance of Modulator. A number of reciprocal crosses of variegated x colorless have been made and the resulting ears are now being grown for comparison of variegation grades. Two other experiments are in progress in which variegated seed was treated with heat (as described by Brawn M.G.C. News Letter 35:83-84) or with acriflavine. Both these treatments are known to "cure" some cells of some cytoplasmic elements. The ears grown from the treated seed will be harvested in a few months' time and if any positive results are obtained, the tests will be repeated on a bigger scale next year.

Nancy van Schaik
Department of Genetics

4. Selection without inbreeding in a South African open-pollinated variety.

An experiment was designed to determine if progress could be made with controlled selection for higher and lower yield in the open-pollinated variety Pretoria Potchefstroom Pearl. The specific aim of the study was to investigate the contribution of additive and nonadditive genetic variation to yield in this variety.

One hundred open-pollinated ears of Pretoria Potch. Pearl were chosen at random and the yield of plants grown from each ear compared. The ten highest and ten lowest yielding lines were selected. A third selection was made of the S_1 progeny of the ten highest yielding S_0 lines. From these, three synthetic selections each consisting of ten families were developed. Each synthetic was tested with Pretoria Potch. Pearl in a yield trial.

The average heritabilities of the High S_0 , Low S_0 , and High S_1 synthetic selections were 0.163, 0.213 and -0.260, respectively.

It may be concluded that little progress can be made using this method in Pretoria Potchefstroom Pearl and that with selection for high yield, interaction plays a more important role in this variety than generally expected, while the contribution of the additive genetic variation is comparatively small.

A. F. du Toit
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Department of Agriculture

5. An indication of genic interaction in maize.

In the local hybrid maize breeding program an effort was made to test all possible crosses of the elite white lines. As these single crosses were tested over a period of four years, each trial contained approximately 30% duplications from other seasons in order to standardize the results.

This method gives rise to some inaccuracy due to gene-environment interaction, but, as the main purpose was a preliminary screening of possible double hybrids, it was felt that the method would be adequate for our needs.

Two complete diallels were compiled from the available data: (1) a 19 x 19 diallel consisting only of lines derived from the variety Pretoria Potchefstroom Pearl and (2) a 23 x 23 diallel containing the above lines together with lines from other sources.

The expected yields of the single crosses according to an additive scheme were calculated as $1/2(\text{average of all the single crosses} + \text{the sum of the average effects of the two parents})$.

If the genic effects are predominantly additive, the distribution of the deviations from the expected yields should not differ significantly from a normal curve. As the deviation from a normal distribution was significant in both cases, P being between .02 and .05 in the first case and smaller than .01 in the second case, it would seem that genic interaction (either intra- or inter-allelic or both) is important in breeding for yield in maize.

J. M. P. Geerthsen
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1. Experiments utilizing radiation in a paramutation program.

A series of experiments have been started to investigate the pre-paramutant and postparamutant effects of radiation. Alterations of the (R^r) paramutant types and the (R^{st} , R^{mb}) types that induce paramutation are being studied. The induction of mutations of R^{st} by gamma and neutron irradiation is also being done and the spectrum of alterations of paramutability will be determined.

A collection of South American races which have variegated aleurones has been made for inclusion in a paramutation program being initiated. The original seeds have been classified for the type of pattern they exhibited using the stippled (R^{st}) and mottled (R^{mb}) types as the code. The following races included seeds of one or both patterns:

Collection	Race	Marbled type (Rmb)	Stippled type (Rst)
Bolivia 491	Altiplano	X	-
Bolivia 663	Altiplano	-	X
Bolivia 905	Altiplano	X	-
Bolivia 591	Huilcaparu	X	X
Bolivia 771	Huilcaparu	X	X
Bolivia 768	Huilcaparu	-	X
Bolivia 876	Huilcaparu	-	X
Bolivia 623	Huilcaparu Moteado	X	X
Bolivia 666	Huilcaparu Moteado	X	X
Bolivia 718	Paru	X	-
Bolivia 724	Paru	X	-
Bolivia 308	Checchi	-	X
Bolivia 320	Checchi	-	X
Bolivia 532	Checchi	X	X
Bolivia 715	Checchi	X	X
Bolivia 833	Checchi	-	X
Bolivia 840	Checchi	-	X
Bolivia 928	Checchi	X	X
Bolivia 454	Cuzco Boliviano	-	X
Bolivia 596	-	X	X
Bolivia 617	-	X	X
Bolivia 621	-	-	X
Bolivia 643	-	X	X
Bolivia 646	-	X	X
Bolivia 648	-	X	X
Bolivia 706	-	X	-
Bolivia 723	-	X	X
Bolivia 733	-	X	-
Bolivia 753	-	X	X
Bolivia 766	-	-	X
Bolivia 967	-	X	X
Chile 434	Capio Grande	-	X
Chile 443	Capio Grande	X	-
Chile 432	-	-	X
Peru 683	-	-	X
Peru 1085	-	-	X
Peru 1094	-	X	X

D. B. Linden

2. Fluorescent compounds in Bf-1.

Isolation and identification of the anthranilic acid-containing blue fluorescent substances in anthers of Bf-1 are being carried out (see MGCNL 32:28, 1958). Extraction of strongly fluorescent fatty substances has been found to improve paper chromatography and chemical fractionation of the blue fluorescent components. The main fluorescent compounds are easily oxidized during purification. One of the substances has been obtained in crystalline form.

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1. Recombination values for 11 alleles at the Wx locus.

Presented here are the recombination values (Wx pollen grains x 10^{-5}) for 54 of the possible 55 F_1 's between 11 wx alleles of independent origin. The results of intercrosses between the alleles C, a, H21, 90, and B have been reported previously. Of the new alleles 1, 2, 4, 6, and 8 were kindly supplied by Dr. R. A. Brink who detected the mutations in an experiment designed to test mutation rate in a P^{VV} stock. After being received at Purdue the mutant alleles were separated from the allele (probably C) in the tester stock used to detect the mutational event. The R allele was furnished by Dr. D. W. Richardson who found the mutation in a stock of popcorn.

The recombinational values for the crosses between the new alleles and between the new alleles and the old alleles are given with considerable reservations. In the first place, the stocks are very heterogeneous with respect to background. We now know that in crosses between two different wx alleles, differences in genetic background can have a pronounced effect upon recombination values. In the second place, data for these new crosses have come from only two plants for each cross. For these reasons, it is felt that the recombination values given may be poor estimates of the frequency of recombination that would be shown by 2 alleles in a common genetic background.

It is felt, however, that a reliable datum for each cross is whether or not any recombination is observed (frequencies below 2×10^{-5} are considered as not showing recombination since some of the parental stocks may have a frequency this high). Table 1 presents the data in both forms for each cross. The order of the alleles is arbitrary as will be shown.

Inspection of the table will show that using lack of recombination as a criterion, one can separate the alleles into three distinct sets. They are (1) C which recombines with every other allele; (2) the set delimited by R and including H21, 4, and 2, none of which recombine with R; and (3) the set delimited by B and including a, 1, 90, 6, and 8, none of which recombine with B. Any member of a set will recombine with all alleles in the other 2 sets. Within a set, 2 alleles may or may not show recombination.

Table 1. Qualitative and quantitative recombination data for 54 F_1 progenies between 11 wx alleles of independent origin. The quantitative data are given as frequencies $\times 10^{-5}$. For the qualitative data, a (0) indicates no recombination observed, a (+) indicates recombination.

	C	a	1	B	90	6	8	4	H21	R	2
C		+	+	+	+	+	+	+	+	+	+
a	5		0	0	0	0	?	+	+	+	+
1	19	0		0	0	0	0	+	+	+	+
B	26	0	0		0	0	0	+	+	+	+
90	80	0	0	0		0	0	+	+	+	+
6	59	0	4	0	0		0	+	+	+	+
8	47	?	6	0	4	0		+	+	+	+
4	49	50	106	48	43	54	112		0	0	+
H21	46	29	49	36	32	71	39	0		0	+
R	61	19	33	32	47	49	40	0	0		0
2	33	46	67	29	58	20	125	61	13	0	

On the basis of recombination or lack of recombination, the alleles can be arranged in a linear array within each set (according to the criterion of Benzer, 1959, P.N.A.S.) although there are too few alleles in any set to make this particularly meaningful. The sets, however, can be arranged in any order with respect to each other and still meet Benzer's specifications.

On the same basis, H21 and 4 are genetically indistinguishable from each other. Either B or 90 are genetically indistinguishable from a (depending on whether or not a should show recombination with 8) but are distinguishable biochemically since a allows limited synthesis of amylose. All other alleles can be shown to be different on this basis which is a rigorous one.

The results emphasize the necessity for having all alleles in a common genetic background if recombination frequencies are to be meaningful. We're meeting this requirement as rapidly as possible.

Oliver Nelson

2. A test for intermediacy of Wx recombinants.

It had been suspected that some Wx alleles arising in crosses between wx⁹⁰ and wx^C or wx^{Coe} were not fully functional. In fact, in the large-scale conventional test of recombination between wx⁹⁰ and wx^{Coe} where Wx kernels were detected by staining, a few Wx kernels were tentatively identified as intermediates because of their weak-staining propensities.

In 1961, 59 viable Wx recombinants from the 1960 test of Bz wx⁹⁰ v / bz wx^{Coe} v were crossed by and onto bz wx^{Coe} v in order to verify their genotypes. The kernels resulting from these crosses were then available to test for amylose percentage.

In the past several months, 20 Wx alleles arising by recombination have been tested for their ability to support amylose synthesis. Only recombinants carrying one or both of the recessive outside markers from the cross Bz wx⁹⁰ v / bz wx^{Coe} v were used in order to be certain that no Wx contaminants were tested. Starch was isolated from the stocks to be tested by the method of Shuman (Report #4 to the Corn Industries Research Foundation, Inc.). Such starch isolates had less than 1% protein as measured by Lowry's assay using the Folin phenol reagent. Protein (gluten) would be the most likely contaminant of any starch sample. The amylose percentage of the starch isolates was measured by the "Blue Value" method given by Ulmann and Augustat (Z. Anal. Chem. 162:337-344, 1958). The "blue value" read in the assay was converted to amylose percentage by reference to an amylose calibration curve constructed from mixtures in varying proportions of 3X recrystallized amylose and amylopectin. Such a standard curve was run with every group of assays.

The results of the assays are given in Table 2. Note that the kernels being tested were either Wx/Wx/wx (Wx recombinant x the bz wx^{Coe} v tester, 15492) or Wx/wx/wx (bz wx^{Coe} v tester x recombinant). The duplicate values given for each cross are assays on different days from the same starch isolate.

Reference to Table 2 shows no definite indication that any of the recombinants can be considered significantly less effective than the check in supporting amylose synthesis. The three recombinants where the reciprocal crosses were analyzed were originally thought to be intermediates because of their weak-staining properties. Even with these recombinants, effectiveness in supporting amylose synthesis cannot be considered less than normal.

It is interesting to observe that our results confirm the observation of Sprague, Brimhall, and Hixon and Sager that the Wx dosage effect on amylose percentage is not linear. However, we derive quite a different dose effect curve. Considering all isolates from Wx/Wx/wx kernels, the mean amylose percentage is 19.8. For all isolates from Wx/wx/wx kernels, the mean amylose percentage is 13.1. Both these values are well below those given by Sprague et al. and Sager.

It is difficult to account for the discrepancy. The assay used here was colorimetric while that used by Sprague *et al* was potentiometric. Yet approximately the same values were obtained for $\underline{Wx}/\underline{Wx}/\underline{Wx}$ isolates. The differences between percentages observed for the lower dosages are large to be attributed to background effect and are most readily ascribable to techniques of measurement.

Table 2. Amylose percentages in starch isolated from the listed stocks.

Source	Genotype	% Amylose
Check	$\underline{Wx}/\underline{Wx}/\underline{Wx}$	27.5, 26.0
"	$\underline{Wx}/\underline{Wx}/\underline{wx}$	20.5, 21.5
"	$\underline{Wx}/\underline{wx}/\underline{wx}$	12.5, 12.5
15424G x 15492	$\underline{Wx}/\underline{Wx}/\underline{wx}$	18.5, 25.5
15492 x 15424G	$\underline{Wx}/\underline{wx}/\underline{wx}$	13.0, 11.5
15425E x 15492	$\underline{Wx}/\underline{Wx}/\underline{wx}$	17.5, 18.5
15492 x 15425E	$\underline{Wx}/\underline{wx}/\underline{wx}$	14.0, 14.5
15427G x 15492	$\underline{Wx}/\underline{Wx}/\underline{wx}$	18.5, 20.0
15492 x 15427G	$\underline{Wx}/\underline{wx}/\underline{wx}$	13.5, 13.5
15422G x 15492	$\underline{Wx}/\underline{Wx}/\underline{wx}$	22.0, 18.5
15422B x "	"	19.5, 20.0
15422D x "	"	21.0, 19.0
15422E x "	"	20.0, 20.0
15422H x "	"	18.0, 17.0
15423A x "	"	18.0, 24.0
15422F x "	"	19.5, 20.0
15423G x "	"	18.5, 19.0
15422C x "	"	19.5, 20.5
15424A x "	"	20.0, 20.5
15423B x "	"	23.0, 20.0
15424B x "	"	24.0, 20.5
15424C x "	"	19.0, 18.5
15424D x "	"	19.5, 20.0
15424H x "	"	19.5, 19.0
15424F x "	"	17.5, 20.0
15425A x "	"	17.5, 20.0

Oliver Nelson

3. The allele \underline{wx}^{m-1} recombines with \underline{wx}^C .

The recombinational pattern of \underline{wx} mutants which apparently contain the requisite information for \underline{Wx} activity but are inhibited in action by a controlling element such as \underline{Ds} would be most interesting to geneticists. We have been endeavoring to make such tests for several years.

In 1960 Dr. Barbara McClintock was good enough to send seed of the heterozygote $\frac{c^{m2} \text{ Sh } wx^{m-1}}{C \text{ sh } wx^S \text{ Ds}}$; Ac. The allele designated by McClintock

as wx^S is almost certainly the same allele which we call wx^C . Plants of this stock were outcrossed to ML4; the F_1 's were selfed in Florida in the winter of 1960-61; from ears on which some of the wx/wx kernels had Wx sectors, non-sectored wx/wx kernels were selected. The plants from these kernels were pollinated by W23 (P^{VV}/P^{VV} ; wx/wx); pollen from these plants was put onto wx^C and wx^{90} ; tassel branches were collected for pollen analysis. Those plants where the Wx frequency in the pollen was less than 2×10^{-5} , which gave kernels with Wx sectors when pollinated by W23 (P^{VV}/P^{VV} ; wx/wx), but which did not induce the formation of kernels with Wx sectors when used as males on wx^C and wx^{90} , were considered to be wx^{m-1}/wx^{m-1} without Ac in the genotype. The crosses of these plants onto wx^C were grown in the first '61-'62 greenhouse crop, and results are given in Table 3. The plants 62G5, 62G8, and 62G12 all come from different isolations of the wx^{m-1} allele. The 62G8A and G8B data came from different plants of the same isolate.

It is apparent that the different wx^{m-1} isolates used in crosses on wx^C show reasonably good agreement as to frequency of Wx pollen grains in the F_1 . The crosses of these plants onto wx^{90} are being grown in the 2nd GH crop, and results will soon be available. Crosses to the other wx alleles will be made in 1962.

It might be of interest to note the frequency of the Wx pollen grains in the heterozygote $\frac{c^{m-2} \text{ Sh } wx^{m-1}}{C \text{ sh } wx^S \text{ Ds}}$ where 1 Ac was present.

These data are given in Table 4. The populations per slide are low since the plants were extremely weak and this apparently reduces pollen production. Where the numbers of Wx pollen grains are as high as these samples, the number of Wx per slide is estimated by the same technique used to estimate the total number of pollen grains per slide.

Table 3. The frequency of Wx pollen grains in the cross $wx^C \times wx^{m-1}$, no Ac.

Plant	Source	Estimated Total Pop.	No. Wx	$Wx \times 10^{-5}$
62G5A	$\frac{15110}{15046-2}$	44,000	7	16
62G8A	$\frac{15102}{15049-2}$	43,500	11	25
62G8B	$\frac{15102}{15049-2}$	55,000	14	25
62G12A	$\frac{15102}{15053-2}$	42,000	8	19

Table 4. The frequency of Wx pollen grains in c^{m-2} Sh wx^{m-1} / C sh wx^C Ds, Ac plants.

Plant	Estimated Total Pop.	No. Wx	Wx x 10 ⁻⁵
12501-1	22,000	536	2436
-2	21,000	320	1524
-3	19,000	609	3205
-4	14,000	402	2871
-5	20,000	93	465
-6	25,000	135	540
-7	9,000	151	1678
-9	18,000	191	1061

Oliver Nelson

4. A test for an unlinked inhibitor of a 5th chromosome gametophyte factor.

Longley (Genetics, 1961) has reported that a 5th chromosome gametophyte factor is one component of a two-component system. The other component is an unlinked inhibitor (In). The Ga gametes have a selective advantage over ga gametes only on the silks of In / - plants (the alleles at the Gametophyte locus in the female plant are immaterial).

Having worked for several years with a 5th chromosome gametophyte factor which is apparently Ga₂, stocks were on hand for a test of whether or not such an inhibitor could be implicated in this particular instance. The stock carrying the Ga factor is an inbred line, 4541, derived from Black Beauty popcorn. This line is A₁, C, R, A₂ Bt Ga Pr. In the F₂ population derived from a cross of 4541 onto Burnham's A₁, C, R, a₂ bt ga pr tester, we have observed a mean bt percentage of 5.1, and an a₂ percentage of 10.4 in a population of 3107 kernels from 7 ears.

The backcross, a₂ bt ga pr / A₂ Bt Ga Pr x a₂ bt ga pr / a₂ bt ga pr and its reciprocal were made in 1960 in order to estimate the recombination between a₂ and bt. Both backcrosses, F₁ x a₂ bt ga pr and a₂ bt ga pr x the F₁, gave proportions of a₂ and bt seeds which were in agreement with the expectation of a 1:1 ratio. The combined estimate (total population = 2457) of a₂ bt recombination was 7.7 percent.

Consider the possible genotypes of the parental strains if a two-component system is applicable. There is a striking deficiency of a₂ and bt kernels in the F₂ progeny. Therefore, 4541 is A₂ Bt Ga Pr and Burnham's tester is a₂ bt ga pr. There is also an In allele contributed by one or both of the parents, but the data from the F₂ progeny are not informative as to this point. However, the backcross a₂ bt ga pr / a₂ bt ga pr x a₂ bt ga pr / A₂ Bt Ga Pr gave .5 bt seeds. Therefore the a₂ bt ga pr / a₂ bt ga pr stock must be in / in since if it were In / In there should be a marked deficiency of a₂ and bt seeds in this backcross progeny. So 4541 must then be A₂ Bt Ga ; In. The backcross onto the tester was a₂ bt ga pr ; in x a₂ bt ga pr / A₂ Bt Ga Pr ; In / in. Since the postulated inhibitor is not linked to the Ga / ga locus,

half the Ga gametes will carry In and the other half in. The same consideration applies to the reciprocal backcross.

From the paired reciprocal BC progenies $a_2 \text{ bt } ga \text{ pr} \times a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr}$ (12722-7 x 12425-6) and $a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr} \times a_2 \text{ bt } ga \text{ pr}$ (12425-6 x 12722-7), seeds which were phenotypically $A_2 \text{ Bt } Pr$ were selected and planted in 1961. Since the Ga factor with which we have been working seems to be about 10 units distal to Bt, the chances that an $A_2 \text{ Bt } Pr$ gamete would carry ga are approximately 1/100 (not taking interference into account) and can be disregarded for predictive purposes. Thus in the plants arising from the $A_2 \text{ Bt } Pr$ seeds from each BC progeny there should be equal numbers of plants which are $a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr}$; In / in (deficiency of a₂, bt, and pr on selfing) and plants which are $a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr}$; in / in (normal ratios of a₂, bt, and pr on selfing).

Thirty plants from $A_2 \text{ Bt } Pr$ seeds from each backcross progeny were grown. Fifteen selfed ears were obtained from the $a_2 \text{ bt } ga \text{ pr} \times a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr}$ progeny and 12 from the reciprocal. The results of scoring these ears for bt and a₂ kernels are presented in Table 1. For each of the 27 ears the numbers of bt and a₂ kernels are significantly less than the 25 percent expected ($P < .05$ for all and $< .01$ for most) if a gametophyte factor were not operative. To find that all 27 progenies show low percentages of bt where one-half are expected to give 25 percent if an unlinked inhibitor In were necessary to give Ga gametes the competitive advantage over ga gametes, suffices to rule out this hypothesis ($P < .001$).

Table 1. Results of selfing $a_2 \text{ bt } ga \text{ pr} / A_2 \text{ Bt } Ga \text{ Pr}$ plants from paired reciprocal BC progenies.

No.	Tester x F ₁ (12722-7 x 12425-6)			F ₁ x Tester (12425-6 x 12722-7)		
	Total	% bt	% a ₂	Total	% bt	% a ₂
- 1	412	4.9	10.9	437	6.9	13.5
- 2	408	3.2	11.0	133	6.0	16.5*
- 3	243	3.7	12.8	162	5.6	8.6
- 4	208	9.6	18.3*	241	8.7	16.6
- 5	222	5.0	12.2	172	15.2	17.4*
- 6	377	4.2	11.7	230	4.8	10.4
- 7	334	5.1	11.7	303	4.0	8.9
- 8	312	6.4	13.1	405	3.7	12.3
- 9	357	6.2	14.0	265	9.8	19.6*
-10	342	3.8	12.9	321	5.6	14.6
-11	400	9.3	18.3	344	5.3	12.5
-12	227	7.5	13.2	332	9.0	11.7
-13	232	7.3	13.4			
-14	233	3.9	11.6			
-15	389	4.6	11.6			

* $P < .05$ that these are drawn from a population that is actually 3+ : 1 bt or a₂. $P < .01$ for all other values.

The demonstration that the results are not those expected if a two-factor system were operative for this Ga factor (which is probably Ga₂), is applicable only to this factor. It shows, however, that not all 5th chromosome gametophyte factors require the presence of an inhibitor (In) at another locus in order to exert their effect.

Oliver Nelson

5. A test for fifth chromosome gametophyte factors in some Mexican races.

The 5th chromosome tester A₁, C, R, a₂ bt pr synthesized by Burnham has been a most useful stock in the investigation of chromosome 5 gametophyte factors because it has not appeared to carry any gametophyte factors itself. For example, in the F₂ of crosses times 4541 (a Black Beauty popcorn inbred which is A₁, C, ²R, A₂ Bt Ga Pr), we have found 5.1 percent bt kernels and 10.4 percent a₂ kernels. Thus, the gametophyte factor in 4541 is approximately 10 C.O. units distal to bt and is apparently at the locus designated by Brieger as Ga₂. With respect to 4541, the tester appears to be ga₂.

The genotype of various Central American races with respect to the 4th chromosome gametophyte factor, Ga₁, has long been of interest to us. Results of various tests have been reported in previous M.N.L.'s. Last year we decided to test a few Mexican varieties for their allelic constitution at the Ga₂ locus by crossing onto the a₂ bt ga pr tester and selfing. Four Mexican races, Celaya, Conico Norteño, Cuatero de la Virgen, and Vandeño were used. The results are recorded in Table 1.

It is evident that for 3 F₂ progenies (those involving Conico Norteño, Cuatero de la Virgen, and Vandeño) there are significantly more bt kernels than expected. There are several possible explanations.

In the first place it is possible that there is a multiple allelic series at the Ga₂ locus such that gametes with a particular allele have a competitive advantage over those gametes carrying an allele which is lower in the series but are at a competitive disadvantage relative to gametes carrying an allele higher in the series. In this particular case, the allele ga₂^B in the Burnham tester stock would be almost completely eliminated when competing against Ga₂ from 4541. It would, however, be at a competitive advantage against the ga₂ alleles from Conico Norteño, Cuatero de la Virgen, and Vandeño. In the case of Cuatero de la Virgen, for example, if the ga₂ locus is approximately 10 C.O. units from bt as we've calculated, fertilization was effected in 68 percent of the ovules by the ga₂^B allele from the tester stock.

Alternatively, it is possible that the Ga₂ locus is not implicated at all in these cases of preferential fertilization but that another gametophyte locus on chromosome 5 is responsible for the excess of bt kernels. We have detected in other stocks the existence of a second gametophyte factor on Chromosome 5. It is located about 30 C.O. units distal to bt. If the a₂ bt ga pr tester were Ga at this locus while Conico Norteño, Vandeño, and Cuatero de la Virgen were ga and if Ga gametes always effected fertilization, then the observed results would be attained. No decision on which alternative is more likely can be made from these data.

Table 1. The totals and percentages of bt kernels in the F₂ progenies of a₂ bt ga pr x various Mexican races (1961).

a) (a ₂ bt ga pr x Celaya)			b) (a ₂ bt ga pr x C.N.)		
Ear No.	Total kernels	% bt	Ear No.	Total kernels	% bt
-1	375	24.5	-1	394	32.0
-2	401	24.7	-2	334	28.1
-3	441	25.9	-3	348	29.3
-4	449	23.8	-4	328	30.8
-5	384	21.4	-5	282	30.5
-6	328	23.8	-6	368	22.8
	<u>2378</u>	<u>24.1</u>		<u>2054</u>	<u>28.9</u>
X ² = 1.13		P > .20	X ² = 16.41		P < .001

c) (a ₂ bt ga pr x G. d.l. Virgen)			d) (a ₂ bt ga pr x Vandeño)		
Ear No.	Total kernels	% bt	Ear No.	Total kernels	% bt
-1	520	30.2	-1	415	37.1
-2	417	30.9	-2	455	27.3
-3	468	34.6	-3	431	29.3
-4	503	30.6	-4	399	28.8
-5	452	28.1	-5	398	28.1
-6	319	37.6	-6	377	30.2
-7	317	32.8			
	<u>2996</u>	<u>31.8</u>		<u>2475</u>	<u>30.1</u>
X ² = 74.08		P < .001	X ² = 34.35		P < .001

Jorge Jimenez T.
Oliver Nelson

SCOTTISH HORTICULTURAL RESEARCH INSTITUTE
Invergowrie, Dundee, Scotland
Department of Genetics

1. Sweet corn investigations.

a. Adaptation at the northern limit of maize cultivation. Sweet corn is the sugary seeded form of maize, and has been grown on a limited scale in Perthshire for some years. It is very sensitive to several external factors, particularly day-length, light intensity, soil temperature during germination and temperature prior to flowering. In addition the crop is readily attacked by the frit-fly (*Oscinella frit.*). Hence the growing of sweet corn in Eastern Scotland, which is at the most northerly limit of its cultivation by Man, affords interesting

opportunities for investigating the problems of adaptation, both environmental and genetical, in an outbreeding species.

The detrimental environmental factors can be controlled to some extent by improving the cultural conditions. Thus sowing in artificial heat reduces the failure to germinate, and use of soil blocks or whale-hide cartons in heated closed frames reduces attacks from frit-flies. Hybrid vigour in maize plays an important role in improving cold tolerance. It also introduces stability, or homeostasis, of the male and female inflorescences under conditions of long days and short nights. Hence a variety to be successful in Scotland must be highly heterotic. The two varieties I previously recommended for growing in England, viz., the F₁ hybrid North Star from New York State and the top-cross Canada Cross, a hybrid of Canada Gold variety with C 13 inbred, were considered most likely to be adaptable to conditions in Eastern Scotland (latitude 56 1/2° N). A "first early" F₁ hybrid Earliking from Minnesota, known to perform well over a wide range of conditions in the northern areas of North America, was also considered possibly to have adaptive potentiality.

It is furthermore recognized that all these strains should have a short maturity range. This means that they must have originated su mutations from the American Northern Flint (su) types and not from the dent types of maize (Cf. Haskell: *Genetica*, 28:308-344).

G. Haskell

b. Selection of a suitable variety for Eastern Scotland.

A trial at Dundee of Canada Cross, North Star and Earliking comprised 4 randomized blocks of paired rows with 10 plants per row, the plot being surrounded by a guard row. Spacing was at 1 ft. between plants and 2 ft. between rows. Seeds were sown singly in whale-hide pots on 16 May, and transferred to open frames on 25 May. Germination was over 96% for all varieties. The crop was planted out on 2 June. A single harvesting was made on 27 September. The percentage yield of marketable ears of each variety was: North Star 75, Canada Cross 68 and Earliking 59 ears. The low yield of Earliking was mainly attributable to severe plant damage from frit fly attack. It is not yet possible to predict a variety's reaction to this pest, which does not attack in North America and so has not been selected against. All the varieties suffered in having relatively poor tassels, but there was just sufficient pollination to provide reasonable seed setting. The quality of Earliking was excellent, and that of Canada Cross very good; but North Star rather lacked sweetness, although its ears were the most uniform.

Open pollinated ears of the earliest and best 8 plants in the plot were saved for seed. These comprised 6 ears from North Star and 1 each from the other two varieties. Their seeds have been mixed to form a new synthetic variety of polycross origin. It represents an initial selection for our local conditions, and it will be further selected in 1962.

G. Haskell

SERVICE DE LA RECHERCHE AGRONOMIQUE ET DE L'ENSEIGNEMENT
Rabat, Morocco

1. Location of fl_2 .

The test results with TB-9b (see News Letter 35:134) have not been confirmed by the study of the progeny, in which the allele Fl_2 reappeared.

On the other hand, a close linkage was found between fl_2 and the gene lazy (la) of chromosome IV. Investigations are being carried on so as to confirm this location.

A. Cornu

2. Inhibiting effects of the gene h_2 on germination and seedling growth.

Investigations on the gene h_2 (see News Letter 32:7) were carried on, particularly on germination and seedling growth.

It was found that the influence of temperature was important on the germinating ability of the seed. The germinating power (% germination after 4 days) as well as the germinating ability (after 7 days' test) of normal and mealy seeds (h) taken from the same ears were measured in four ovens set at 10° C, 15° C, 20° C and 25° C respectively. The following results were obtained:

	Germinating power		Germinating ability	
	+	h	+	h
10° C	0	0	0	0
15° C	60	6	100	44
20° C	94	28	100	52
25° C	100	66	100	72

This test shows that germination of the h -seed is much slower than that of the normal seed. Their total germinating ability varies according to temperature, whereas that of the normal seed remains the same at temperatures ranging from 15° C to 25° C.

Moreover, the germinating ability of the h -seed decreases rapidly. After one year's time, it drops from 44 to 16% at 15° C while it stays at 100% for the control lot of the normal seed.

The growth of seedlings in water also shows great differences between normal and (h h) individuals. After a period where development is parallel in both groups of individuals (a six days' period corresponding to seed soaking and germination), the two curves move apart and, on the twelfth day, the green weight of the normal seedlings is about twice that of the (h h) ones.

These inhibiting effects vary, besides, with the more or less homozygous character of the strains used. The effects are less important, or even nil, when heterozygosis is more pronounced.

A. Cornu

3. Double mutant stocks with mealy endosperm.

Starting with h_2 and other known mutants that modify the endosperm structure, double recessive stocks have been constituted in order to study the influence of these factors on one another. Stocks now available are: $h_1 h_2$, $f_1 h_2$ and $sh_1 h_2$. The building of stock $o_2 h_2$ is under way.

A. Cornu

TENNESSEE AGRICULTURAL EXPERIMENT STATION
and
UNITED STATES DEPARTMENT OF AGRICULTURE
Knoxville, Tennessee

1. Gamete deletion in male-sterile crosses.

Previous reports (Agron. Abs., 1959, p. 60, and M.N.L. 1960) have shown that a greater than expected number of fertile plants is obtained when pollen segregating for Rf and rf is used to pollinate plants carrying 33-16(J) male-sterile cytoplasm. Excesses of fertile plants have also been obtained in certain crosses involving Texas (T) male-sterile cytoplasm. It was suggested (M.N.L. 1960) that the excess of fertile plants could be caused by differential competitive effects between Rf and rf pollen grains such that genotypes carrying rf are eliminated. Several studies the past two years support this suggestion.

Pollinating plants carrying T cytoplasm with equal quantities of pollen from T331, a nonrestoring inbred, and Ky21, restoring, produced 80.3 percent fertile plants while pollinating plants carrying J cytoplasm with equal quantities of pollen from Ky27, a nonrestoring inbred, and 33-16, restoring, produced 67.8 percent fertile plants.

In another experiment, plants with T cytoplasm were pollinated with nonrestoring pollen and 1/2, 1, 2, and 4 hours later were pollinated with restoring pollen. The following numbers and percentages of fertile and sterile plants were obtained from these pollinations:

	Fertile		Sterile	
	No.	%	No.	%
Tcms x $rf\ rf$	0	0	300	100.0
" x " + $Rf\ Rf$ 1/2 hr. later	1151	96.7	39	3.3
" x " " 1 " "	347	72.3	133	27.7
" x " " 2 " "	307	58.6	217	41.4
" x " " 4 " "	89	20.1	354	79.9

Ears pollinated with nonrestoring pollen only produced full seed set. Also the delayed pollinations gave similar results from the base, middle, and tip portions of the ears. The results obtained, therefore, could not be due to delayed silking.

These experiments indicate that Rf pollen grains germinate faster than rf grains, pollen tubes from Rf pollen grains grow faster than rf tubes, or both.

Some backcrosses made with F_1 pollen in J cytoplasm have failed to segregate sterile plants. Pollen abortion in these crosses may occur following the four-spore stage. The only evidence bearing on this possibility is that these tassels have had a trace to 50 percent aborted pollen grains at time of shed.

L. M. Josephson

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Genetics Foundation

1. Synapsis and crossing over in plants hyperploid for *Tripsacum* chromosome material.

Unexpectedly high trivalent frequencies have been found in certain 21 chromosome stocks containing a pair of *Zea-Tripsacum* interchange chromosomes. The constitutions of these plants have been described in detail elsewhere (Maguire 1961, Exp. Cell Res. 24:21-36). The chiasma frequencies inferred from these trivalents have been such as to suggest that crossover frequency (between homologous corn segments) was increased from an expected 60% (30 crossover units) to close to 100% in the distal half of the short arm of chromosome 2 in this material. Stocks of appropriate genetic constitution for simultaneous tests of recombination frequencies in the ws_3 , lg_1 , gl_2 region and parental trivalent frequencies have been constructed, and data should be available for analysis during the 1962 season.

In addition material containing these interchange chromosomes was irradiated with the hope of recovering new chromosomal constitutions in which synapsis, chiasma frequency and recombination frequency can be studied. Two such new constitutions are currently being cytologically examined. Other new constitutions are available this season from rare crossovers between corn and *Tripsacum* segments in 20 chromosome plants followed by backcrosses to 21 chromosome plants. It is thought that a series of types of constitution will provide quantitative information on pairing relationships and on unusual chiasma frequency as a function of unusual material present.

Marjorie Maguire

2. An additional common locus in corn and *Tripsacum*.

In previous studies a portion of a *Tripsacum* chromosome which had been substituted for the distal half of the short arm of corn chromosome 2 was found to carry dominant alleles for lg_1 and gl_2 . Recent tests have indicated the presence of a normal allele for ws_3 also on the *Tripsacum*-derived segment. Synapsis at pachytene appears normal between the similar *Tripsacum* and *Zea* segments in plants in which no closer pairing partners are available, but crossing over apparently occurs between them only rarely.

Marjorie Maguire

3. Variability in length and arm ratio of pachytene chromosomes.

Absolute length measurements of all chromosomes from each of 271 pachytene microsporocytes from 89 anthers of 31 related corn plants have been made in the course of various studies over a period of two years. Arm ratios were calculated where centromeres could be identified. These data are listed below:

Chromosome	Mean length (μ)	Standard deviation	Mean arm ratio	No. observations of arm ratio	Standard deviation
1	83.5	19.4	1.33	52	.200
2	69.2	14.9	1.42	56	.251
3	64.5	14.9	2.16	48	.558
4	59.8	12.6	1.59	73	.259
5	58.4	12.9	1.16	56	.145
6	48.3	12.0	3.10	20	.841
7	49.6	10.8	2.83	57	.587
8	46.3	10.1	3.06	52	.584
9	44.5	10.7	1.86	63	.375
10	37.0	8.8	2.70	55	.498

Statistical studies indicate that longer chromosome arms have greater variances but smaller coefficients of variability and that chromosomes with larger arm ratios have inherently more variable arm ratios. These greater variabilities in arm ratio are not correlated with corresponding greater variabilities in length of the chromosomes involved. The results are consistent with the interpretation that the chromosomes have two kinds of variability: one which seems to contribute approximately uniformly per unit length to variability throughout the genome while the other kind of variability may be a characteristic property of each chromosome unrelated to length in any consistent way. Chromosome knobs do not appear to influence variability in length of chromosome arms in which they occur. *Tripsacum* chromosome material does not appear to differ in its length variability from corresponding corn chromosome material for which it has been substituted.

Marjorie Maguire

4. Further studies on pachytene pairing failure.

Pairing failure in pachytene chromosomes was studied against a genetically constant background. Very significant negative correlations were found between total chromosome length per cell on the one hand and number of terminal pairing failures and their total extent (percent length) on the other, and between number of terminal failures and their average extent (percent length). No significant correlation was found, however, between total chromosome length per cell and the average extent (percent length) of pairing failure. If the pairing failures are dissociations increasing in extent and number during the pachytene stage studied, the simplest reconciliation of the results requires that the average rate of their extension be roughly proportional to total chromosome length or that certain constants be related to each other in specific ways. An alternate interpretation, that the pairing failures were present before pachytene and that chromosomes

shorten faster in cells containing them, is favored by the finding that heterogeneity is low between anthers in number of failures per cell and that no significant correlation was found between anthers in total chromosome length and number of pairing failures. The possibility also exists that the pairing failures are a complex combination of both initial synaptic failure and later dissociation.

Distributions of chromosomes containing pairing failure at both ends and of those containing both terminal and intercalary pairing failures generally follow expectations of randomness.

Marjorie Maguire

TEXAS AGRICULTURAL AND MECHANICAL COLLEGE
College Station, Texas

1. Fourth cycle reciprocal recurrent selection results.

Yield tests involving 4th cycle selections crossed on the appropriate composites were grown in 1961. Coefficients of Variation were 9.1 and 11.1 percent and yields were good. Composites and crosses among composites were compared also. Definite progress has been made in each cycle in increasing the yielding ability of the group of top crosses involving the Ferguson's Yellow Dent Selections x Yellow Surcropper testers or composites. The same trend was obtained by using as a check, either the Yellow Surcropper Variety or the mean of two Texas hybrids. In the other group, Yellow Surcropper selections x Ferguson's Yellow Dent testers or composites, progress has not been as consistent. Apparently the third cycle results were influenced by poor stands and unusual weather conditions to such an extent that selection was not very effective. This group of top crosses was grown a year later than the third cycle of the other group. However, if the first, second and fourth cycle results are compared, a steady shift toward higher yielding top crosses has occurred also in this group.

Mean yields of crosses among varieties and composites grown at two locations in 1961.

Crosses among testers	College	Temple
	Station	
	bu. per acre	bu. per acre
YS variety x FYD composites	71.2	58.0
YS ₁ composite x FYD composites	74.1	64.2
YS ₂ composite x FYD composites	79.6	65.7
YS ₃ composite x FYD composites	73.1	61.1

FYD variety x YS composites	73.0	62.5
FYD ₁ composite x YS composites	76.7	62.6
FYD ₂ composite x YS composites	74.8	62.0
FYD ₃ composite x YS composites	73.4	62.1
C. V.	15.5%	9.4%

Actual yields of varieties and composites at two locations in 1961.

Variety or composite	College Station	Temple
	bu. per acre	bu. per acre
YS variety	48.5	46.0
YS ₁ composite	61.9	51.3
YS ₂ composite	68.8	52.0
YS ₃ composite	61.7	52.0

FYD variety	49.5	51.2
FYD ₁ composite	52.4	53.9
FYD ₂ composite	57.0	54.7
FYD ₃ composite	64.6	60.7

In both groups, the lower-yielding top crosses have been reduced in each cycle. Also variation among top crosses was reduced in the fourth cycle tests.

Yields of crosses among testers in composites indicate that a large portion of the increased combining ability can be attributed to the YS composites, especially the YS₂ composite. The accompanying table shows no change in the combining ability of the FYD composites. Actual yields of composites may indicate different types of gene action in the two source varieties.

J. W. Collier

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Plant Industry Station

1. In 1961 several thousand seeds from a cross (B14 x 4063) x A C R B Pl were germinated in the dark and classified for purple root color to identify monoploids. In addition to the expected monoploids, a class of plants was found which were of normal fertility and presumably diploids. These came from kernels having colored aleurone and the plants lacked purple color. In every case such plants, when selfed, were found to be heterozygous for yellow endosperm color. The parental single cross was (Y x y). Therefore the exceptional class of plants is interpreted as being maternal diploids. Maternal diploids and monoploids occurred with roughly equal frequency.

G. F. Sprague

2. An F₂ three-point test involving Bt Pr gl₁₀ / bt pr Gl₁₀ gave the following results:

Bt Pr Gl	Bt Pr gl	bt Pr Gl	bt Pr gl	Bt pr Gl	Bt pr gl	bt pr Gl	bt pr gl
306	161	28	7	98	0	39	1

Crossover percentages are higher than normal for the Bt-Pr and Pr-Gl regions but the suggested three-point order is Bt-Pr-Gl. My stocks of gl₈ and gl₁₀ have either become mixed or these 2 glossies are identical.

G. F. Sprague

WASHINGTON UNIVERSITY
St. Louis, Missouri

1. Genetics of tillering.

A project has been initiated to investigate what if any genetic basis exists for tillering in some of the races such as Parker's Flint in contrast with many midwest inbreds and other races such as Zapalote Chico. Crosses have been made this winter in Florida between Parker's Flint and a series of translocations compiled by E. G. Anderson and E. B. Patterson. In addition, tillering and non-tillering sibling plants from several backgrounds were selfed in 1961; these seeds will be grown and the progeny checked further this coming year.

It is the aim of this project to try to relate expressed morphology more closely to genetic background. Since environment plays a seemingly significant role in tiller development, knowing whether or not specific genes for tillering exist in a particular plant should provide a means of separating environmental and genetic influences on tiller and presumably other aspects of plant development with greater accuracy than is now possible. The aid of E. G. Anderson, E. B. Patterson and W. L. Brown has been enlisted in various ways.

N. H. Nickerson

2. Responses of certain tassel and dwarfing genes to growth substances.

A series of tests was run during 1961 on na₁, na₂, py, br₁, cr₁, d₁, d₂, nl, rt, ba, Cg, and Tp. Groups of plants were subjected to one of the following treatments: Gibberellic acid, naphthalene acetic acid, indole butyric acid, GA-NAA, GA-IBA. Treatments were applied in several concentrations at two-day intervals. NAA stimulated root growth and stalk stiffness, but decreased branching, stature, leaf size, inflorescence size and fertility. IBA stimulated overall growth, but not height; leaves were wider, often longer, more tillers developed, they produced functional inflorescences and root growth was enhanced. GA retarded root growth and inhibited development of lateral branches. In a few cases, it increased height. Generally, effects of IBA-GA or NAA-GA were additive. br₁ / br₁ was markedly affected by GA; leaves were only 1/3 as wide as controls, plants were shorter and with culms 1/2 as great in diameter and inflorescences did not develop. nl / nl plants remained nl; ba / ba plants remained barren. Stature was only slightly modified in na₁ and na₂ plants. rt plants formed roots when treated with IBA or NAA. Tp and Cg plants behaved under effects of GA treatment as previously published; with IBA their increase in vigor was marked, while with NAA they showed few growth differences from controls.

Studies are continuing this year on effects of these and other growth substances alone and in combination on the above-listed and other mutant forms. The purpose of such investigations is to ascertain whether or not effects of specific genes which differ from their alleles in normally-growing plants can be modified or overcome by applied substances which have been found to influence plant growth. The most clear-cut example is still the overcoming of \underline{d}_1 by GA discovered by Phinney. Another which may be equally clear-cut is overcoming of \underline{rt} by auxins, noted above. Suggestions as to genes which could be tested will be welcome.

N. H. Nickerson
R. E. Lindahl

3. Responses of stalk and tassel mutants to TIBA.

Tri-iodo benzoic acid (TIBA) is a synthetic substance which has been shown to cause loss of polar movement of auxin (IAA). During 1961, groups of \underline{ts}_1 , \underline{ts}_2 , \underline{ts}_4 , \underline{Ts}_5 , \underline{Ts}_6 , \underline{sk} , \underline{Cg} , \underline{Tp} and \underline{la} plants were subjected to daily treatments of either dist. H_2O , 100 μg , 500 μg or 1000 μg of TIBA from 2 weeks of age until tassel emergence. In general, doses below 500 μg were ineffective in causing growth changes. An exception was $\underline{sk} / \underline{sk}$, where doses of 100 μg inhibited brace root development entirely and resulted in greatly foreshortened plants; $\underline{sk} / \underline{sk}$ plants treated with higher doses died. In $\underline{ts}_1 / \underline{ts}_1$ plants, main shoots were killed and 2 tillers developed, each with \underline{ts}_1 tassels. Plants were also 1/2 height of controls. $\underline{ts}_2 / \underline{ts}_2$ plants were reduced in height by the higher concentrations but were essentially unchanged in tassel appearance. $\underline{ts}_4 / \underline{ts}_4$ plants were slightly increased in height by 100/ μg doses; the effect was more pronounced on $\underline{+} / \underline{ts}_4$ plants. \underline{Ts}_5 and \underline{Ts}_6 plants were shortened by higher concentrations, but they did not die. 1000 μg doses caused a general chlorosis and often death of tips of leaves as well as general inhibition of brace root development. They also apparently prevented normal cell differentiation in some strains; stalks were of smaller diameter and far more flexible than controls. Tiller production of \underline{Cg} and \underline{Tp} was not strongly affected, but a lowering in height over controls was common. $\underline{la} / \underline{la}$ plants did not remain upright, but fell over from lack of roots rather than from the ageotropic growth characteristic of control $\underline{la} / \underline{la}$ plants. These studies are being continued.

N. H. Nickerson
M. T. Shealey

4. Effects of high concentrations of auxins on normal maize plants under field conditions.

Field-grown plants of the hybrid Spancross showed no detectable growth responses to season-long daily treatments with NAA, IBA and IAA in concentrations ranging from 10^{-8} up to 10^{-3} , the effective ranges of auxin activity employed in laboratory experiments. In preliminary trials of concentrations somewhat higher than these levels, detectable growth effects were obtained. Studies are continuing to find out the limits of tolerance of these substances and their effects on growth of other inbreds, races and hybrids.

P. R. Kremer
N. H. Nickerson

5. Responses of Sorghum plants to gibberellic acid.

Treatment of dwarf, medium and tall varieties of Sorghum with GA resulted in excessively thin and almost non-productive plants at concentrations of 125 and 625 μg GA every 3rd day. However, an amount of 25 μg caused, in addition to the usual suppression of tillers, development of inflorescences two weeks earlier than controls; plants were also shorter by 15 cm, 30 cm and 60 cm than corresponding controls. Studies are continuing, employing stocks which insofar as possible differ only in the number of dwarfing genes present.

N. H. Nickerson
T. N. Embler

UNIVERSITY OF WISCONSIN
Madison, Wisconsin

1. Paramutagenic action of Navajo mutants from a presumed stippled-Navajo compound allele.

The occurrence of a presumed stippled-Navajo compound allele, symbolized as $\underline{R}^{st:nj}$, among the offspring from an $\underline{R}^{st} / \underline{R}^{nj} / \underline{r}^{g\phi}$ x $\underline{R}^r \underline{R}^r$ mating was reported in News Letter 34 (1960). This allele mutates rather frequently to a stable form giving the Navajo phenotype. Unlike ordinary Navajo, however, which is non-paramutagenic, the Navajo mutants from the compound are paramutagenic in heterozygotes with \underline{R}^r at about the same level as the parent allele, as the following data show.

Genotype testcrossed on $\underline{r}^g \underline{r}^g \phi\phi$	Pigmentation score of $\underline{R}^r \underline{r}^g \underline{r}^g$ testcross kernels $\underline{/1}$
$\underline{R}^r \underline{r}^g$ (control)	5.48
$\underline{R}^r \underline{R}^{st}$ (control)	2.32
$\underline{R}^r \underline{R}^{st:nj}$	3.36
$\underline{R}^r \underline{R}^{nj-1}$ (mutant from $\underline{R}^{st:nj}$)	3.33
$\underline{R}^r \underline{R}^{nj-2}$ (mutant from $\underline{R}^{st:nj}$)	3.16
$\underline{R}^r \underline{R}^{nj-3}$ (mutant from $\underline{R}^{st:nj}$)	3.77
$\underline{R}^r \underline{R}^{nj-4}$ (mutant from $\underline{R}^{st:nj}$)	3.14

$\underline{/1}$ 1 = colorless, 7 = self-colored.

It appears probable that these Navajo mutants result from change of the stippled component of the compound to self-color, thus permitting the Navajo component to express itself in the endosperm in the usual form. Retention by the mutants of paramutagenic action about equal to

that of the parent allele finds a parallel in McWhirter's earlier observation that many, but not all, self-colored mutants from ordinary R^{st} are still strongly paramutagenic in $R^r R^{st}$ heterozygotes.

R. A. Brink

2. A phenotypic comparison of three stippled alleles.

Three stippled alleles (R^{st}) have been compared on the basis of aleurone-pigmenting effects, dosage effects, and interaction with the stippled modifier, M^{st} .

The stippled alleles were:

R^{st-1} -- from Wisconsin genetic stocks,

R^{st-2} -- from Maize Co-op. stocks,

R^{st-4} -- a "mutant" originally found heterozygous with R^{sc80} (a self-coloured mutant) in an exceptional plant in the progeny derived by self-pollination of a plant of $R^{sc80} R^r$ genotype. There is circumstantial evidence for origin of R^{st-4} by mutation of R^{sc80} , but recurrence of the mutation was not obtained.

The stippled alleles were incorporated in W22 inbred background, and matings were made among stocks carrying R^{st} , $R^{st}M^{st}$, r^rM^{st} and r^r , to obtain the endosperm genotypes required. The data reported are from the matings which enable an analysis of the dosage effect of the stippled alleles in absence of the modifier, and the dosage effect of the stippled modifier when stippled is held constant at 1 dose.

The number of pigmented spots, in an area enclosed by a 10 x 10 reticule grid at 30x magnification, on the abgerminal face of the kernel was measured. This area was approximately 6 mm². The mean scores reported are based on 125 kernels (25 from each of five ears) for each endosperm genotype with the exception of combinations 1-1 and 1-3 for R^{st-4} . The latter means were based on 100 kernels (25 kernels from each of four ears).

The first three columns of the table show the aleurone-pigmenting effect and dosage response of the three stippled alleles, in the absence of the stippled modifier. The three stippled alleles differ markedly in the frequency of self-coloured spots at each of the dosages 1-0, 2-0 and 3-0. R^{st-1} produced a linear increase in frequency of pigmented spots with increasing dosage. R^{st-2} and R^{st-4} were non-linear in dosage effect. $R^{st-2} r^r r^r$ kernels (combination 1-0) were essentially colourless, only one of 125 kernels examined had a pigmented aleurone spot.

The interaction of M^{st} with the stippled alleles is shown by the comparison of the columns headed 1-0 with 1-1, and 2-0 with 2-1 for each of the R^{st} alleles. Substitution of M^{st} for m^{st} resulted in marked increases in the frequency of pigmented spots. Interaction with M^{st} may be held to be an objective criterion for distinguishing stippled alleles, and all three alleles showed the interaction. The distinctive effects of each of the stippled alleles are maintained in these combinations, however, as is shown by cross comparison between R^{st} alleles.

The dosage effect of \underline{M}^{st} , when the dosage of the stippled allele is held constant at one dose ($\underline{R}^{st} \underline{r}^r \underline{r}^r$ kernels), is shown by the columns headed 1-1, 1-2 and 1-3. With each of the stippled alleles an increase in \underline{M}^{st} dosage was attended by an increase in frequency of pigmented spots. \underline{R}^{st-4} showed a linear increase in frequency of pigmented spots with increasing dosage of \underline{M}^{st} , while \underline{R}^{st-1} and \underline{R}^{st-2} showed a non-linear response to dosage of \underline{M}^{st} .

These data show that the three stippled alleles have distinctive phenotypic effects, and may be further differentiated by characteristic dosage effects, and response to increasing dosages of the specific stippled modifier.

Table 1. Mean number of pigmented spots per kernel, in a defined area, for the endosperm genotypes involving combinations of stippled alleles and the stippled modifier.

Stippled allele	Dosage of stippled and \underline{M}^{st} (1)						
	1-0	2-0	3-0	1-1	1-2	1-3	2-1
\underline{R}^{st-1}	6.3 ±0.17	20.3 ±2.36	37.9 ±1.44	15.4 ±0.86	48.5 ±2.94	47.2 ±1.60	61.6 ±4.08
\underline{R}^{st-2}	0.008 ±0.08	6.2 ±0.43	7.7 ±1.64	5.5 ⁽²⁾ ±0.80	27.8 ±0.20	34.4 ⁽³⁾ ±1.37	41.6 ±2.86
\underline{R}^{st-4}	0.3 ±0.04	0.9 ±0.15	4.4 ±0.29	9.3 ±0.91	15.3 ±0.99	20.7 ±2.51	17.8 ±1.21

(1) The first digit represents the number of stippled alleles in the triploid endosperm, the alternative being \underline{r}^r . The second digit represents the number of \underline{M}^{st} elements, the alternative being \underline{m}^{st} .

(2) Constitution of these kernels was $\underline{R}^{st-2}\underline{M}^{st-2}/\underline{r}^r/\underline{r}^r$.

(3) Constitution of these kernels was $\underline{R}^{st-2}\underline{M}^{st-2}/\underline{r}^r\underline{M}^{st-1}/\underline{r}^r\underline{M}^{st-1}$, all other combinations involved the indicated stippled alleles with \underline{M}^{st-1} (extracted from $\underline{R}^{st-1}\underline{M}^{st-1}$).

K. S. McWhirter

3. Paramutability of \underline{r}^r mutants from standard \underline{R}^r .

The standard \underline{R}^r allele, which was first observed to undergo paramutation in heterozygotes with \underline{R}^{st} , mutates most frequently to either of two types of alleles which are complementary in phenotype, \underline{R}^g (colored aleurone and colorless, or green, seedlings) and \underline{r}^r (colorless aleurone and red seedlings) (Brink; Quart. Rev. Biol. 35:120-137, 1960). Eight \underline{R}^g mutants from standard \underline{R}^r were found to be indistinguishable from the parent allele in aleurone pigmentation action and in paramutability in heterozygotes with \underline{R}^{st} . (Brink et al., Gen. 45:1297-1312; 1960). The analogous comparisons between the standard \underline{R}^r allele and its derived

\underline{r}^r mutants require the measurement of pigmentation in vegetative seedling tissues and are technically more difficult to make. A further criterion for paramutation at the \underline{R} locus was suggested by the discovery that \underline{R}^r and \underline{R}^g alleles which have undergone paramutation in heterozygotes with \underline{R}^{st} have also become weakly paramutagenic. (Brown and Brink; Gen. 45:1313-1316, 1960). Consequently, nine \underline{r}^r mutant genes were tested for paramutagenic action following heterozygosity with \underline{R}^{st} .

The paramutagenic action of each of the various alleles was tested, after two generations of heterozygosity with \underline{R}^{st} , in heterozygotes with \underline{R}^g_2 , a mutant from standard \underline{R}^r . The following crosses were made:

(1)	$\underline{r}^r \underline{R}^{st}$	}	x	$\underline{R}^g_2 \underline{R}^g_2$
(2)	$\underline{R}^r \underline{R}^{st}$			
(3)	$\underline{R}^r \underline{r}^r$			
(4)	$\underline{r}^g \underline{r}^g$	x	$\underline{r}^{r'} \underline{R}^g_2$	
(5)	$\underline{r}^g \underline{r}^g$	x	$\underline{r}^r \underline{R}^g_2$	
(6)	$\underline{r}^g \underline{r}^g$	x	$\underline{R}^{r'} \underline{R}^g_2$	
(7)	$\underline{r}^g \underline{r}^g$	x	$\underline{R}^r \underline{R}^g_2$	

$\underline{R}^{r'}$ and $\underline{r}^{r'}$ genes (extracted from heterozygotes with \underline{R}^{st}) and the corresponding control \underline{R}^r and \underline{r}^r genes (with no history of heterozygosity with \underline{R}^{st}) were combined with \underline{R}^g_2 in crosses (1) to (3). Crosses (4) to (7) are testcrosses of the resulting $\underline{r}^{r'} \underline{R}^g_2$, $\underline{r}^r \underline{R}^g_2$, $\underline{R}^{r'} \underline{R}^g_2$, and $\underline{R}^r \underline{R}^g_2$ progeny.

A comparison of the aleurone phenotypes of the $\underline{R}^g \underline{r}^g \underline{r}^g$ kernels from crosses (4) and (5) constitutes a test for paramutagenic action of the $\underline{r}^{r'}$ alleles. A similar comparison between crosses (6) and (7) provides a measure of the paramutagenic action of $\underline{R}^{r'}$ under comparable conditions.

Fifty $\underline{R}^g \underline{r}^g \underline{r}^g$ kernels from each testcross ear were scored against a standard set of kernels defining seven pigmentation classes ranging from colorless (class 1) to self colored (class 7). The data obtained are summarized in Table 1. Each mean score entered in the table is derived from testcrosses of ten plants.

The mean scores entered in the bottom line of the table show that the pigmenting capacity of \underline{R}^g_2 has been weakened in heterozygotes with $\underline{R}^{r'}$. The difference between the scores for the $\underline{R}^g \underline{r}^g \underline{r}^g$ testcross kernels from the two classes of staminate parents is significant at the .01 level of probability. There is no indication of a similar loss in pigmenting action of \underline{R}^g following association with any of the $\underline{r}^{r'}$ alleles, as shown in the remainder of the table. The mean scores for the $\underline{R}^g \underline{r}^g \underline{r}^g$ testcross kernels from the $\underline{r}^{r'} \underline{R}^g$ staminate parents are actually higher than the scores of those from $\underline{r}^r \underline{R}^g$ staminate parents for seven of the nine mutants. The difference is statistically significant (at the .05 level) only in the case of \underline{r}^r 40. The basis for this apparent enhancement of \underline{R}^g action is not clear at the present time, but a similar effect has been noted incidentally in \underline{Rr} plants in other pedigrees not involving the \underline{R}^{st} allele. An evaluation of the significance of the effect must await further study.

Table 1. Mean aleurone color scores for $\underline{R}^{\underline{G}}\underline{r}^{\underline{G}}$ kernels from the crosses $\underline{r}^{\underline{R}}\underline{r}^{\underline{G}} \times \underline{r}^{\underline{R}'}\underline{R}^{\underline{G}}$ and $\underline{r}^{\underline{G}}\underline{r}^{\underline{G}} \times \underline{r}^{\underline{R}}\underline{R}^{\underline{G}}$. The bottom line gives the corresponding scores from the crosses $\underline{r}^{\underline{G}}\underline{r}^{\underline{G}} \times \underline{R}^{\underline{R}'}\underline{R}^{\underline{G}}$ and $\underline{r}^{\underline{G}}\underline{r}^{\underline{G}} \times \underline{R}^{\underline{R}}\underline{R}^{\underline{G}}$.

Allele Tested	♂ Testcross parent		P*
	$\underline{r}^{\underline{R}}\underline{R}^{\underline{G}}$ (or $\underline{R}^{\underline{R}}\underline{R}^{\underline{G}}$)	$\underline{r}^{\underline{R}'}\underline{R}^{\underline{G}}$ (or $\underline{R}^{\underline{R}'}\underline{R}^{\underline{G}}$)	
$\underline{r}^{\underline{R}}$ 30	5.54	5.49	>.5
$\underline{r}^{\underline{R}}$ 31	5.73	5.67	>.5
$\underline{r}^{\underline{R}}$ 32	5.54	5.68	.1 > P > .05
$\underline{r}^{\underline{R}}$ 33	5.45	5.55	.3 > P > .2
$\underline{r}^{\underline{R}}$ 34	5.41	5.54	.1 > P > .05
$\underline{r}^{\underline{R}}$ 37	5.56	5.77	.1 > P > .05
$\underline{r}^{\underline{R}}$ 38	5.64	5.80	.1 > P > .05
$\underline{r}^{\underline{R}}$ 39	5.67	5.74	>.5
$\underline{r}^{\underline{R}}$ 40	5.49	5.73	.02 > P > .01
$\underline{R}^{\underline{R}}$	5.29	5.02	.01 > P > .001

*P is the probability of the null hypothesis by the t test.

It is clear from the data summarized in Table 1 that the $\underline{r}^{\underline{R}}$ mutants, while heterozygous with $\underline{R}^{\underline{S}t}$ for two generations, have not acquired the capacity to induce paramutation of $\underline{R}^{\underline{G}}$ to a more weakly pigmenting form, whereas the parent $\underline{R}^{\underline{R}}$ allele has. The mutational events by which the $\underline{r}^{\underline{R}}$ alleles arose have, in each case, altered the capacity of the \underline{R} locus to undergo paramutation in heterozygotes with $\underline{R}^{\underline{S}t}$. The $\underline{r}^{\underline{R}}$ mutants differ from the $\underline{R}^{\underline{G}}$ mutants, both derived from standard $\underline{R}^{\underline{R}}$, in this respect.

Douglas Brown

Addendum:

ATOMIC ENERGY ESTABLISHMENT TROMBAY
Bombay, India
Biology Division

1. Pigmented silk scar.

A pericarp variant in maize, characterized by the development of red pigmentation around the silk scar, has been repeatedly collected from the local Bombay markets. When crossed with either P^{RR} or P^{WW} (or even when self-pollinated), the original phenotype is seldom recovered. The P^{RR} allele is apparently extracted unaltered. On the other hand, when crossed (after selfing for one generation) with a highly inbred homozygous coloured aleurone stock (kindly supplied by Prof. R. A. Brink), it disclosed an unusual property of inhibiting the aleurone pigmentation. Thus, (1) when crossed as the male parent, the resulting kernels were considerably weaker in pigmentation, and (2) when mated as the female parent, the resultant kernels with a few exceptions were completely colourless. This property seems somewhat similar to that of Greenblatt's Diffuse (MNL 33:129-130), wherein Df/df pollen placed on silks of A C R plants produces 5-10% smoky kernels, with the remainder self-coloured. In the present case, however, pigmentation in all the kernels seems to be weakened, although the intensity of pigmentation is not uniform.

Table 1

Types and numbers of kernels obtained from backcrossing the F_1 between Red Silk Scar Pericarp and a colored aleurone stock, Wis. $A_1 A_2 C R$, as female parent to the colored aleurone stock.

Cob no.	Pedigree no.	Genotype	Aleurone pigmentation grades					Total	Color index
			Full color- ed	Color- less					
				5	4	3	2		
1	$\frac{B60-33-21}{29-12A}$	Red Silk Scar Pericarp / Wis. $A_1 A_2 C R$ Wis. $A_1 A_2 C R$	36	27	27	32	16	138	3.25
2	$\frac{B60-33-30}{29-21}$	"	8	5	4	4	6	27	3.18
3	$\frac{B60-35-2}{29-12B}$	Wis. $A_1 A_2 C R$ / Red Silk Scar Pericarp Wis. $A_1 A_2 C R$	38	8	8	13	16	83	3.47

Table 2

Types and numbers of kernels obtained on selfing the F_1 between Red Silk Scar Pericarp and a colored aleurone stock (Wis $A_1 A_2 C R$).

Cob no.	Pedigree no.	Genotype	Aleurone pigmentation grades					Total	Color index	
			Full colored		Colorless					
			5	4	3	2	1			
1	B60-33-23 (X)	Red Silk Scar Pericarp (X) Wis. $A_1 A_2 C R$	--	--	--	--	298	298	1.0	
2	B60-33-25 (X)	"	21	15	11	5	111	163	1.9	
3	B60-33-26 (X)	"	--	--	--	--	114	114	1.0	
4	B60-33-27 (X)	"	31	14	9	14	107	175	2.1	
5	B60-33-30 (X)	"	50	13	32	10	149	254	2.2	
6	B60-34-1 (X)	"	56	17	25	21	209	328	2.05	
7	B60-34-2 (X)	"	13	2	4	2	56	77	1.9	
8	B60-34-3 (X)	"	32	11	12	7	189	251	1.8	
9	B60-34-4 (X)	"	33	21	30	14	180	278	2.0	
10	B60-34-5 (X)	"	44	20	20	20	233	337	1.84	
11	B60-34-6 (X)	"	61	31	15	42	169	318	2.2	
12	B60-34-7 (X)	"	65	29	22	21	144	281	2.4	
13	B60-34-8 (X)	"	53	24	21	36	142	276	1.9	
14	B60-34-9 (X)	"	35	34	22	37	191	319	2.01	
15	B60-35-1 (X)	Wis. $A_1 A_2 C R$ Red Silk Scar Pericarp (X)	83	4	15	46	153	300	2.4	
16	B60-35-6 (X)	"	50	23	19	18	207	317	2.02	
17	B60-35-9 (X)	"	34	9	7	9	68	127	2.4	
18	B60-35-10 (X)	"	53	21	20	25	131	250	2.3	
19	B60-33-29 (X)	Red Pericarp								
			Average color index:							
Wis. $A_1 A_2 C R$ Red Silk Scar Pericarp			= 2.28					Red Silk Scar Pericarp / Wis. $A_1 A_2 C R$ = 1.88		

Backcross of the F_1 with the colored aleurone stock as the recurrent male parent yielded kernels giving a whole spectrum of aleurone pigmentation ranging from completely colored to completely colorless. Kernels were scored against a set of standards of 5 intensities of pigmentation. The data are given in Table 1. If a simple partially dominant inhibitor is postulated to explain the F_1 data, the backcross data show a preponderance of colored kernels (when all colored grades are lumped together). Various two factor hypotheses also do not seem to reconcile the F_1 and backcross data.

The F_2 data are recorded in Table 2. Attention is drawn to the fact that 2 cobs gave only colorless kernels. Secondly, the average score of pigmentation for the crosses $\frac{A \ C \ R}{\text{Pigmented Silk Scar}} \textcircled{x}$ is somewhat higher than that of the crosses $\frac{\text{Pigmented Silk Scar}}{\text{Wis. A C R}} \textcircled{x}$.

The differences, however, are not statistically significant.

N. K. Notani
Chandra Mouli

2. Extreme modifications in radiosensitivity of maize seeds stabilized for different moisture contents.

Maize seeds of inbred Oh57, when irradiated with gamma radiation doses of 10 to 50 Kr. following their stabilization for moisture contents ranging from 1.87 to 10.55%, showed extreme modifications of radiosensitivity as measured by seedling height and survival. The radiation sensitivity varied by a factor of about 5. The maximum differential in radiosensitivity was attained at 10 Kr. In contrast, hulled barley seeds of a local variety, stabilized similarly for their moisture content (1.97 to 12.01%) and irradiated with the same doses gave a differential in radiosensitivity of a factor of less than 2. Furthermore, the maximum differential was obtained at the dose of 40 Kr. Barley seeds, in addition, showed an increase in radiosensitivity at the highest moisture level (12.01%). It was inferred that when the full range of moisture contents of the seeds is examined, the seeds show an increase in radiosensitivity both at low and very high moisture levels.

N. K. Notani
B. K. Gaur

III. STOCKS AVAILABLE AND WANTED

A. Wanted:

W. D. Bell, Pennsylvania State University:
Chlorophyll-deficient mutants other than complete albinos,
particularly yellow-stripes, green-stripes, pale green or
yellow seedlings.

B. Available:

W. D. Bell and J. E. Wright, Pennsylvania State University:
After this growing season, chlorophyll-deficient mutants for
physiological study.

C. R. Burnham, University of Minnesota:
(Ra Ra) gl₁ v₅
Multiple recessive bm pr ys virescent with expanded glumes.

W. C. Galinat, Harvard University:
OP seed from an F₁ hybrid of A158 gl₃ X T. floridanum.
Seed of T. floridanum, T. dactyloides 2n of Kansas and
T. dactyloides 4n of Florida.

IV. REPORT ON MAIZE COOPERATIVE

The procedure of routinely converting our genetic stock collection toward one or more of the inbred lines M14, W23, and Oh51A has been discontinued. The single cross hybrid W23 X L317 is now being used in this program for deriving vigorous, adapted stocks.

The volume of seed requests continues to rise each year. The increase in requests from abroad has been especially striking, and it is now probable that more than half the seed distributed each year is sent to foreign countries. Requests from physiologists and biochemists have been building up gradually for several years. Demand has been heaviest for traits affecting starch composition of the endosperm and for dwarfs and chlorophyll mutants. A special seed increase of traits in these categories was made this past summer.

Dr. Johnie Jenkins, who had been assisting part-time with the work of the Maize Cooperative, joined the staff of the U.S.D.A. Boll Weevil Research Laboratory, Starkville, Mississippi, last July 1.

Dr. E. G. Anderson currently has a half-time appointment on our staff as Visiting Professor of Genetics. He is spending part of the year at Pasadena sorting out stocks from his collection for transfer to the Maize Cooperative.

Requests for stocks or for copies of stock lists should be sent to the Botany Department, University of Illinois, Urbana, Illinois.

The following listing of reciprocal translocations represents a supplement to last year's catalog of stocks. The interchange positions for these translocations are available from the following source: Longley, A. E., Breakage Points for Four Corn Translocation Series and Other Corn Chromosome Aberrations. U.S. Dept. of Agr., Agr. Res. Serv. ARS 34-16, 40 pp., 1961.

RECIPROCAL TRANSLOCATIONS

<u>Symbol</u>	<u>Translocation</u>	<u>Symbol</u>	<u>Translocation</u>
8001	1-9	8219	5-6
8004	4-8	8249	1-4
8006	3-7	8302	1-9
8023	3-8	8321	2-5
8027	2-4	8322	2-7
8032	3-9	8339	4-6
8041	1-5	8345	5-10
8045	2-7	8347	1-5
8048	1-3	8349	3-10
8069	4-5	8350	3-8
8103	4-7	8351	3-5
8104	3-5	8367	3-8
8108	4-5	8368	1-4
8143	6-7	8374	4-7
8219	2-10	8375	1-10

<u>Symbol</u>	<u>Translocation</u>	<u>Symbol</u>	<u>Translocation</u>
8376	2-8	8649	4-9
8380	4-6	8651	6-10
8383	7-9	8658	1-6
8386	5-9	8659	7-9
8388	1-5	8662	2-3
8389	1-9	8663	1-4
8395	4-5	8665	5-6
8397	3-4	8666	} 3-8
8405	1-3	8667	
8407	2-4	8670	
8412	3-10	8671	5-7
8415	1-6	8672	3-6
8420	5-8	8679	5-7
8428	4-6	8683	1-8
8439	6-9	8696	5-6
8441	2-6	8746	5-8
8443	3-4	8764	4-6
8447	3-9	8768	6-9
8452	1-6	8770	1-10
8457	5-9	8782	1-5
8460	1-9	8786	2-6
8465	3-9	8796	5-8
8483	2-3	8806	5-8
8491	1-10	8818	5-6
8525	8-9	8854	5-9
8528	3-5	8864	2-10
8536	6-9	8886	} 1-9
8541	4-10	8890	
8558	7-9	8895	5-9
8562	3-9	8904	6-10
8563	1-4	8906	6-9
8580	7-8	8919	1-8
8590	5-6	8927	4-6
8591	5-9	8951	8-9
8591	4-6	8963	3-6
8602	1-4	8972	1-5
8607	4-8	8987	4-8
8609	1-6	8995	1-3
8622	4-5	8997	5-8
8628	1-2	9002	2-6
8634	3-4	9020	8-10
8636	4-9	9028	4-10
8637	1-3	48-34-2	1-5
8640	1-8	48-40-8	4-7
8645	6-10		

<u>Symbol</u>	<u>Translocation</u>	<u>Symbol</u>	<u>Translocation</u>
001-3	1-10	015-9	1-10
001-5	8-10	015-10	5-9
001-13	1-8	016-15	7-8
001-15	3-7	016-17	3-6
001-15	2-6	017-3	1-2
002-12	4-5	017-18	2-4
002-16	2-5	018-3	2-4
002-17	5-8	018-4	4-5
002-19	1-4	018-5	1-5
003-5	2-8	018-18	1-2
003-16	4-6	019-1	2-5
004-3	7-8	019-3	7-10
004-7	3-7	020-5	3-9
004-7	4-9	020-7	5-9
004-11	1-2	020-19	1-8
004-13	2-4	021-1	7-8
004-17	5-6	021-3	4-5
005-7	1-8	021-5	4-10
005-14	2-3	022-4	2-7
006-7	4-5	022-11	5-9
006-10	2-8	022-15	7-10
006-11	5-10	022-20	5-10
006-17	3-4	023-2	2-3
007-17	5-8	023-5	2-3
007-19	1-10	023-13	5-7
008-17	1-8	023-15	2-5
008-18	5-9	024-1	6-8
009-19	2-5	024-5	1-5
010-4	2-4	024-7	1-9
010-10	2-3	024-11	3-8
010-12	1-7	024-14	1-3
011-7	2-4	024-16	4-10
011-11	6-7	025-4	2-5
011-16	4-6	025-12	4-6
011-20	2-8	026-2	1-8
012-16	3-4	027-4	2-6
013-3	5-7	027-6	6-7
013-8	6-7	027-9	7-9
013-9	1-3	027-10	4-5
013-11	5-8		
013-17	2-8		
014-5	5-8		
014-12	2-3		
014-17	7-8		
015-3	2-5		

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