test crosses of plants 6452-3, 6453-3, and 6453-9 deviated very much from 1:1 and with plant 6453-9, marked differences in ratio of these two phenotypes were expressed on ears produced from different parts of this plant. Nevertheless, it was clear that some chromosomal component, present in the $a_1 m^{-1}$-carrying plants and not linked to this locus, was segregating at meiosis and that the expression of a uniformly pale class of kernels and a class showing deeply pigmented spots in a colorless background was related to the presence or the absence of this component. Since in the grandparent (5719A-1), in the two $a_1 m^{-1}$ parent plants, and in all plants of cultures 6452 and 6453, variegation was expressed, it seemed evident that the presence of this component in the chromosome complement was related to variegation and its absence to the appearance of the pale phenotype.

As stated earlier, none of the kernels on the ears derived from test crosses with the $a_1 m^{-1}$-carrying plants grown in the summer of 1951 had been examined carefully up to this time. When it was realized that the ratio of kernel types appearing on the ears of plants entered in table 3 segregation of an independently located element in the chromosome complement associated with control of $a_1$ behavior, the kernels on ears produced by test crosses of plants in the parent culture, 6040.
were then examined. More direct evidence of the presence of this element was obtained, for it was found to be linked in some plants to a genetic marker, \( y \), in chromosome 6 (\( Y \), yellow starch in endosperm; \( y \), recessive allele, white starch in endosperm). The plants in this culture were derived from kernels on an ear produced by the cross of a plant homozygous for \( a_1, Sh_2 \) and \( y \), plant 5719A-1, which was \( a_1^{-m-1} Sh_2/a_1 Sh_2, Y/y \). Some kernels in the variegated \( Y \) class on this ear were sown in the summer of 1951 under A of culture 6047. Others that were variegated \( Y \) were sown under B and C of culture 6047. In addition, variegated kernels that were \( Y \) were selected from the ear produced by self-pollination of plant (\( A \), table 2) 5719A-1 and sown under B and C of culture 6046. All plants in cultures 6046 and 6047 received some type of test by means of crosses to plants of known constitution, by self-pollination, or both. The kernel types on ears obtained by crosses of these plants to plants that were \( a_1, Sh_2 \) and \( y \) are given in table 4. All plants in this table were \( a_1^{-m-1} Sh_2/a_1 Sh_2 \). Those in Part I were \( Y/y \) whereas those in Part II were \( y/y \).

In Part I of table 4, a ratio of 1 pale to 1 variegated kernel appeared among those showing pigment was exhibited among the kernels on ears produced from crosses conducted with plants 6046B-1 and 6047A-3.
Linkage of the pale phenotype with Y and the variegated phenotype with y is clearly expressed by the test with plant 6046B-1. In plant 6047A-3, these linkage relations were reversed; the pale phenotype was linked with y and the variegated phenotype with Y. The ratio of kernel types obtained from tests of the other plants in Part I of Table 4 indicated that each carried more than one controlling element in the system. In plants 6046B-3 and 6046C-2, evidence of one of these elements with Y was evident but in plant 6047A-1, no evidence of such linkage was shown. The plants in Part II of Table 4 were all y/y in constitution and thus no linkage relationships could be shown by them. Nevertheless, the evidence suggested the presence of more than one independently located controlling element in all plants except 6047C-3. The plants in culture 6452 (Table 3) originated from the y class of variegated kernels produced by the cross of plant 6047A-3 entered in Table 4. According to the data of Table 4, this plant had only one independent controlling element and it was located in the Y carrying chromosome. The plants in culture 6453 originated from the variegated class of kernels produced from the test cross of plant 6047C-4, Table 4. The ratio of kernel types on its test cross ear suggested the presence in this plant of two independent controlling elements, each at a different location in
the chromosome complement. Selection of the class of variegated kernels was made in order that the color of the anthocyanin pigment in the aleurone layer in kernels on ears of plants derived from them would be expressed on a white rather than a deep-yellow background. As stated earlier, the only purpose of this test had been to determine crossing over between $a_1^{m-1}$ and $Sh_2$.

From the ratio of kernel types given in table 4, and from those obtained from self-pollination and from the test cross of plant 5719A-1 ($A$ and $C$ of table 2), it would appear that plant 5719A-1 had more than one independently located controlling element, and that one of them occupied a position in its $Y$ bearing chromosome 6.

As shown above, the plants in culture 6452 (table 3) arose from variegated kernels on the ear of plant 6047A-3, whose kernel types are entered in table 4. These plants would be expected to have only one independently located controlling element. This appeared to be true for all plants except plant 3. The plants in culture 6453, table 3, arose from variegated kernels on the ear produced by plant 6047C-3, whose kernel types are entered in table 4. These plants would therefore be expected to have either one or two of the independently located controlling elements. As table 3 indicates, seven of the nine plants in this culture had one such element.
but the remaining 2, plant 3 and plant 9, had more than one, such element and the ratio of kernel types on ears produced by different parts of plant 9 suggested that the number of this element was not the same in all parts of the plant. It was suspected from this that the independently located element could undergo transposition in somatic cells and that this was responsible for the observed deviation from expected ratios.

3. Initial evidence suggesting the primary mode of operation of the independently located controlling element in the \( a_{m-1} \) system, the suppressor-mutator.

The mode of control of \( a_{m-1} \) action induced by the independently located element of the system, whose discovery was outlined above, was not appreciated until the results of tests made in the summer of 1953 were analysed. From them, a reasonable interpretation could be drawn of its mode of operation. With this in mind, all test cross ears that had been produced in the summer of 1951 were then carefully examined. The evidence obtained from them fitted nicely with this interpretation.

It was possible then, to derive this interpretation. This suggested the types of precise test of this that should be made and these were conducted in the summer of 1954. Before these tests are considered, the evidence leading to an appreciation of the mode of operation of the independently located controlling element will be outlined, briefly.
Due to unforeseen conditions, the total planting during the summer of 1953 had to be restricted. Space was available for only a fraction of the number of plants usually grown in the summer season. For this reason, study of $a_{1}^{m-1}$ was limited, preference being given to other projects then under investigation. The reason that the decision was made to limit study of $a_{1}^{m-1}$ was dictated by the fact that up to this time, many of the testcross ears obtained from tests conducted in the summer of 1951 had not yet been examined and little was really known of the behavior of $a_{1}^{m-1}$. It was decided, then, to limit its study to an examination of the position of the independently located element in the system in the progeny obtained from plant 6047C-2 (table 1), and to the plants derived from the pale class in the progeny of plant 6046C-4.

This latter plant had been derived from a variegated kernel on the self-pollinated ear of plant 5719A-1 (table 2). The constitution was $a_{1}^{m-1} Sh_{2}/a_{1}^{m-1} Sh_{2}, Y/y, pr/pr$. The ear from which the pale kernels were selected had been produced by self-pollination. It gave rise to 406 kernels, with the following phenotypes: 35/pale colored kernels of which 33 were $Y$ and 2 were $y$ and to 371 kernels that had deeply-pigmented spots in a colorless background and 267 of these were $Y$ and 104 were $y$. Twenty pale kernels were selected from this ear and plants grown from them under
The decision to select pale kernels from this ear was because the plant was homozygous for $a_{m-1}$ and thus each chromosome should carry a derivative of $a_{m-1}$. It was desired to examine the behavior of each of these derivatives.

Therefore the plants derived from the pale kernels were tested for this by crosses with plants homozygous for $a_1, sh_2$ and $y$. Plant 6047C-2, table 4, had the constitution $a_{m-1} \text{Sh}_2 a_1 \text{Sh}_2, \text{pr/pr}$, and $Y/y$. An ear of this plant had been crossed by that was homozygous for $a_1, sh_2, y$ and had $\text{Pr}$ in one chromosome 5 and pr in its homologue. The types of kernels appearing on the resulting ear, given in table 4, suggested that plant 6047C-2 had one of the independently located elements of the $a_1$ system in $Y$ bearing chromosome and, in addition, two others located elsewhere in the chromosome complement. Ten kernels in the variegated, $Y, \text{Pr}$ class and ten other in the variegated, $y, \text{Pr}$ class were selected and sown in the summer of 1953 under culture number 6629A and B. All the ears produced by the plants derived from these kernels were used in crosses with plants that were homozygous for $a_1, sh_2$ and $y$. Two stocks were used for this, but their constitution with respect to the Pr alleles was not then known. Tests conducted with them indicated that one stock was $\text{Pr}/\text{Pr}$ and the other $\text{pr}/\text{pr}$. The need for knowing the constitution with respect to $\text{Pr}$ alleles is
will be evident shortly. A few ears of plants in culture 6629 were either self-pollinated or sib crossed.

The phenotypes of kernels on the ears of plants in culture 6629A, produced by the test cross, are entered in table 5. Again, it will be noted that there is very close linkage between the kernels carrying the Sh2 marker and those exhibiting pigment. For each plant, the position of the ear on the plant is indicated. It will be noted that on the ears of plants A-2, A-5, and A-8, and on the first ear of the main stalk of plant A-7, there was a ratio of pale kernel to variegated kernel among the anthocyanin pigment bearing classes, and that the pale phenotype was linked with Y whereas the variegated phenotype was linked with y. To facilitate ready appreciation of this, the phenotypes of the pigment bearing classes of kernels on these ears are entered in table 6. On the remaining ears in table 5, a more ready appreciation of exhibited other ratios, and for the numbers of kernels in the pale and variegated classes are entered in table 7. It would appear from these ratios that more than one independently located controlling element was present in plants A-2, A-5 and A-8. Linkage of the pale class with Y was given by the ratio of the pale class of kernels on the ear obtained from plant A-8 but on none of the other ears was there good evidence of this. The ratio of kernel types on the ear produced by the tiller of plant A-7 was very aberrant, only 4

The position of the ear on the plant is indicated. It will be noted that on the ears of plants A-2, A-5, and A-8, and on the first ear of the main stalk of plant A-7, there was a ratio of pale kernel to variegated kernel among the anthocyanin pigment bearing classes, and that the pale phenotype was linked with Y whereas the variegated phenotype was linked with y. To facilitate ready appreciation of this, the phenotypes of the pigment bearing classes of kernels on these ears are entered in table 6. On the remaining ears in table 5, exhibited other ratios, and for the numbers of kernels in the pale and variegated classes are entered in table 7. It would appear from these ratios that more than one independently located controlling element was present in plants A-2, A-5 and A-8. Linkage of the pale class with Y was given by the ratio of the pale class of kernels on the ear obtained from plant A-8 but on none of the other ears was there good evidence of this. The ratio of kernel types on the ear produced by the tiller of plant A-7 was very aberrant, only 4
variegated kernels appeared among the total of 112 that had anthocyanin in
them. From the ratio of Sh$_2$ to sh$_2$ on this ear, it was clear that there
was no deficiency in transmission of the a$_1$ bearing chromosome.

The types of kernels appearing on the ears produced by plants in B
culture 6629, following the test cross with plants that were homozygous
for a$_1$, sh$_2$ and y, are entered in table 6. An approximate 1:1 ratio
of pale to variegated kernels was expressed on the ears produced by plants
B-1, B-3, B-4, B-7, and B-10 and on the first ear of B-5. On the remaining
10 ears, whose kernel types are entered in this table, other ratios of these
two classes of kernels were expressed. On several ears, a class of kernels
appeared that exhibited a marked alteration in pattern of variegation.
The number of such kernels is indicated by an asterisk. These kernels
were colorless except for 1, 2, or several very small dots that exhibited
the A$_1$ phenotype. It will be noted, also, that on those ears in which they
appeared in appreciable numbers, likewise exhibited a
marked increase in the proportion of the colorless Sh$_2$ class of kernels.
As will be shown later, these totally colorless, Sh$_2$ kernels, with rare
exceptions, belong with the variegated class of kernels. On a few of the
ears whose kernel types are entered in table 5, one or several kernels
exhibiting only one or a very few small dots of the A$_1$-type pigment likewise
were present but because the table was complicated, they were not indicated in it, and a description of them was deferred until this time. (It may be stated now, however, that study of the progeny derived from these kernels with the very decided modification of variegation expression revealed the Spm-w state of the Spm element and this will be considered in detail in section 00.) It should be mentioned also that the kernels exhibiting the modified variegation pattern were not always randomly distributed over the ear. On some ears, they were adjacently aligned, forming a cluster on the ear.

In addition to the test crosses conducted with plants in culture 6629, already described, an ear of a tiller of plant 6629A-2 and 6629A-4 self-pollinated and pollen collected from this tiller had been placed on the silks of a tiller ear of plant 6629B-2 and 6629B-6. The ratio of kernel types on the resulting ears need not be given here as they add little to the information already gained. However, the phenotypes of some kernels among them served to illucidate the mode of operation of the independently located element and this will be discussed shortly after the test crosses with plants in culture 6628 are considered.

As mentioned earlier, page 00, the plants in culture 6628 were derived from the pale, Y, pr class of kernels on the self-pollinated ear of plant
which was homozygous for $a_1^{m-1}$, $Sh_2$ and $pr$ but heterozygous for the alleles of $Y$ ($Y/y$). In order to learn whether the derivatives of $a_1^{m-1}$ in each chromosome 3 would express the pale phenotype and in the same manner by each, these plants were crossed by plants homozygous for $a_1$, $sh_2$ and $y$.

All kernels on the resulting ears were uniformly pigmented and the grade of intensity of this was the same in all of them. Some of the $a_1$, $sh_2$, $Y$ tester plants were homozygous for $Pr$ whereas others were homozygous for $pr$. From comparisons of pigment type in kernels on ears produced by use of these two types of tester plant, it was clearly apparent that with $Pr$ present, an intense anthocyanin pigmentation appeared in the aleurone layer of the kernel, resembling close to that appearing when $A_1$ is present.

The intensity of anthocyanin pigment in kernels that were homozygous for $pr$ was much less than that which is produced with $A_1$. There would be no confusion in identifying a pale, $pr$, from the $A_1$, $pr$ class. With this in mind, attention will be given to the note entered at the base of table 5 and table 8.

Here is given the number of kernels that expressed the full $A_1$ phenotype but only mixtures in the $pr$ class. Those of this type in the $Pr$ class could not always be distinguished from the easily and therefore they are included in the pale classes in these tables. The frequency of appearance of kernels with an $A_1$ phenotype is so low that inclusion of a few of them in the pale
classes does not invalidate the significance of the given ratios in these tables. \(\text{[In this regard, it may be mentioned here that in subsequent study of} \ a_1^{m-1} \text{it was often necessary to use another state of} \ a_1^{m-1} \text{that gives} \)

rise to kernels exhibiting a pale phenotype in both the purple (Pr) and red (pr) classes that is readily distinguished from the purple and red classes produced by \(A_1\).)

In examining the kernels on the ears derived from cross of plants 6629B-2 and 6629B-6, which were \(y/y\), by plant 6629A-4, which was \(Y/y\), some kernels appeared that had several areas exhibiting the pale phenotype.

In the Pr class, these areas were quite intense. In the pr class, in contrast, the pigment intensity in these areas was low. In fact, with respect to pigment intensity, the pale areas were similar to those expressed by the pale kernels in the Pr and pr classes entered in tables 5 and 8. In plant 6629A-4, one independently located controlling element was present and carried in its \(y\)-bearing chromosome. By means of crossing over, some of the \(Y\) carrying gametes produced by this plant should have this element, and in the crosses with plants in 6629B, some kernels on the resulting ear should have only one of this element and carrying the element.
Among the Y class of variegated kernels on this ear, several were found that exhibited white starch immediately beneath a pale area, and the correspondence in their borders was exact. There seemed little doubt that the convenient expression of pale area and white starch, a normally-creeping type of appearance of the pale area, was associated with loss of the segment-bearing 6 of the X chromosome that removed both Y and the independently located controlling element. In other words, the pale areas appearing in the variegated kernels arose from mutation or removal (or inactivation) in some somatic nuclei. Among these elements resulted an expression of the pale area that was associated with loss of the segment-bearing 6 of the X chromosome that removed both Y and the independently located controlling element. This could be concluded from the observation that the pale areas appearing in the variegated kernels arose from mutation or removal (or inactivation) in some somatic nuclei. Among these elements resulted an expression of the pale area that was associated with loss of the segment-bearing 6 of the X chromosome that removed both Y and the independently located controlling element.

No attempt was made to cross plants of culture 6629 to those of culture 6628, which were homozygous for a derivative of a(m-1) and for pr, and y. Similar types of cross had been made in the summer of 1951 even though the ears derived from them had not been examined. However, on one ear of a plant of culture 6628, which had been used in a cross with a plant homozygous for a1, a2, Y, and pr, a single variegated kernel was present. The plant was homozygous for a1, a2, Y, and pr. It was a contaminant in that it had Pr and Y instead of pr and y. It was obviously derived from functioning of a stray pollen grain produced by a plant in culture 6629A. These plants were growing in the next row and were immediately adjacent to those in culture 6628. Since this kernel showed spots of deep pigment in a colorless background as well as a few pale areas, it seemed evident that the independently located element
in the a<sub>m-l</sub> system had a dual function. In its absence, a<sub>m-l</sub> functioned in the production of anthocyanin pigment, although in a manner not totally comparable to A<sub>1</sub>. In the presence of this element, all evidence of this function was suppressed. In its absence, the mode of functioning of a<sub>m-l</sub> was constant, no change in this occurred. In its presence, on the other hand, return to the A<sub>1</sub>-type functional capacity of action was initiated and A<sub>1</sub>-type expression arose in some cells, and the time during development when this change in mode of action occurred in a cell, and the number of cells in which this took place, was genetically controlled. Careful examination of ears produced during the summer of 1951 was then immediately undertaken to gain enough insight into the mechanism associated with control of gene action at a<sub>m-l</sub> had been gained to allow meaningful interpretations to be drawn from examinations. Up to this time, this had not been true. These examinations substantiated the interpretation of the mode of operation of the independent element and therefore this element was given the designation Suppressor-mutator and symbolized as Spm. Before considering the evidence obtained from these examinations, a resume will be given of the results so far obtained, the interpretations drawn from them, and the problems they posed.
Studies outlined in this and the previous section were conducted with the progeny of one plant, that of plant 5719A-1. In this plant, the expression of the \( a_1^{m-l} \) locus, carried in one of its chromosomes, was very different from that given by the parent plant having \( a_1^{m-l} \). As stated in section 3 of this report, it was considered to have an altered state of \( a_1^{m-l} \). In plants having this state and also Spm, small streaks with deep-anthocyanin pigmentation appear in a non-pigmented background although occasionally, a large area exhibiting this phenotype may appear.

In the kernel, small deeply pigmented spots appear in a non-pigmented background (photo. 1) although, here also, occasionally, a large area of this type appears. The described \( a_1^{m-l} \) phenotype was exhibited by the kernel that gave rise to plant 5719A-1, by the plant itself and it also appeared in the majority of variegated plants and kernels in the progeny of 5719A-1 that were carried through three generations. However, on some test cross ears produced by some of the progeny plants, a few kernels appeared that exhibited a marked deviation from this pattern. In them, the number of deeply-pigmented spots was very much reduced or none appeared. No explanation for this had yet been found. Also, on a few ears produced by the test cross, a kernel exhibiting the full \( A_1 \)-type pigmentation appeared. The nature of the
change at \( a_{-1} \) responsible for their appearance had not yet been
determined, although it was suspected to come from inbreeding of
that strain. The gene action was probably in the expression.
In the absence of Spm, the 5719A-1 state of \( a_{-1} \) gave rise to
plants that were uniformly pigmented but the intensity of this, and its
distribution within the plant differed from that expressed when \( a_{-1} \) is
present. The kernels were likewise pigmented in the absence of Spm
and this pigment was uniformly distributed over the aleurone layer of the
kernel. It was intense when \( Pr \) was present but light when the
kernels were homozygous for \( pr \). The phenotype appeared to be quite
stable and reappeared unaltered in progeny of plants exhibiting it if
Spm was absent, these plants were either self-pollinated or crossed by plants in the
tester stocks that were homozygous for \( a_{-1} \).

From the ratio of pale to variegated kernels on the ears produced
the
by/test cross of plant 5719A-1 (C, table 2), and from the ratio of these
(kernel types on ears produced by test crosses of its progeny, it was
concluded that plant 5719A-1 had at least three Spm elements in it and
that one of them was located in its \( y \) bearing chromosome. However, the
number of this element in the progeny derived from a test cross did not
always conform with that expected, and the test of plant 6452-3, table 3,
illustrates this. It was expected to have one Spm element but the ratio of
pale to variegated kernels deviated markedly from the expected one to one.

Also, similar types of test conducted with ears on different parts of the
same plant or with pollen from a plant, sometimes produced of these two classes a
ratio in the resulting ears that did not agree with one another.

This suggested that the Spm constitution in different parts of an
individual plant were not alike. It was suspected that these differences
arose from somatically occurring transposition of Spm, but no tests of this
had yet been conducted.

As emphasized above, the interpretations of the Spm system
were drawn from evidence obtained only from this one isolate of aI. Others that
produced quite different phenotypes had been isolated, it
was therefore necessary to determine to what degree the behavior of each of
them would conform with the interpretation that had evolved from study of
but since all tests had been conducted with them during the summer of 1951
and when the ears produced by these tests were examined, it was realized
that the interpretation would apply to them. It was considered that
each isolate of aI, originally selected because of the modified
variegation pattern it had exhibited, arose from some alteration that had
occurred to the original aI, and that each such alteration was responsible
for the type of variegation pattern exhibited in the presence of Spm and
also for the type of expression given in its absence. Some of the
evidence for this now be reviewed.

Additional evidence supporting the interpretation of the primary

Substantiation of the mode of action of the primary

individuality of states of $a_{m-1}$.

During the summer of 1951, plants were grown from selected kernels
that were homozygous for $a_1$. Fifty-two plants were derived from kernels
exhibiting the pattern of variegation produced by this original state of

one $a_{m-1}$ (photo. ). Seven were derived from kernels whose aleurone layer
was uniformly pigmented, but the intensity of this was low. In addition,
plants were grown from two kernels, with very much modified patterns of
variegation. One had very many small, deeply pigmented spots in a colorless
background. The other had many such spots and, in addition, several
large, deeply pigmented areas. In addition, kernels were selected from
ears produced by crosses of some of the plants grown in the greenhouse
during the winter of 1950-51. Selections were made from ears produced
by self-pollination or by a cross with a plant homozygous for $a_1$,
either in the progeny $a_{m-1}$ of the plant having $a_{m-1}$.

These kernels selected from these plants to be grown in the summer of 1951
were as follows: plants 5700A, 5718, 5719A-1, 5719A-2, and 5720. The ratio
of kernel types on ears produced by plants 5718, 5719A-1, and 5719A-2
are described below. Table of the plants derived from these will then be
are entered in table 2.
Plant 5700A, derived from a very pale colored kernel, developed intense anthocyanin pigment in stalk, leaf-sheath, and glumes. It was used in a cross with a plant homozygous for $a_1$, and on the resulting ear, half of the kernels were very pale colored and half were colorless.

This plant carried a derivative of $a_1^{m-1}$ that had been received from the original $a_1^{m-1}$ plant, in one of its chromosomes 3 and $a_1$ in the other. Plants were grown from some of the pale kernels on this ear.

From the self-pollinated ear of plant 5718, some of the variegated kernels were selected and plants grown from them. Plant 5720, described in section 3, had a modified state of $a_1^{m-1}$ in one of its chromosomes 3 and $a_1$ in the other. The modified state produced both large and small areas in plant and kernel that exhibited only low levels of pigment intensity in them. Variegated kernels from the several ears produced by crosses were sown in the summer of 1951.

On the self-pollinated ear of plant 5719A-1 and of plant 5719A-2, (A, table 2), and on ears produced by cross of these plants to plants homozygous for $a_1$, (C, table 1), some kernels appeared that had areas of various sizes and colors of pigment similar to that appearing in the variegated kernels exhibiting a pale phenotype in addition to the deeply-pigmented spots which appeared in all variegated kernels. The number of such kernels among the variegated class was low. The majority of them exhibited only...
deeply-pigmented spots that were nearly always small in size. Therefore, many both types of variegated kernels on ears produced by self-pollination and by the tests and plants were grown from each type of variegated kernel.

Table 9 is constructed to show the types of kernels that were selected from ears produced by crosses of plants grown in the greenhouse, and the culture number given to the plants derived from each selection.

Discussion will commence with tests conducted with plants derived from selected kernels on the ear produced by the cross of plant 5719A-2 with one that was homozygous for \( a_1 \). Plant 5719A-2 was \( a_1^{-m-1}/a_1 \) in \( (C, V_{a_1}^{1/2}) \) constitution. On the ear this cross produced, there were 24 uniformly pale colored kernels, 250 kernels exhibiting dots of deep/color in a colorless background, and 259 totally colorless kernels. Among the variegated class of kernels, some showed, in addition to the deeply-pigmented dots, areas of light pigmentation, all of which had the same intensity. In a few kernels, these lightly pigmented areas were large, covering as much as an eighth of the area of the alabrone layer. There was, however, much variation in size of such areas within a single kernel. Some of the kernels exhibiting these areas were selected and the plants grown from them given culture number 6081A.

The frequency of appearance of different classes of kernels on the ears of plants homozygous and heterozygous in respect to self-pollinated ears, on ears produced by crosses with plants homozygous
appearing on ears produced by test crosses with similar types of test conducted with the parent plant, 5719A-2 (A, B, C, D, table 2). There was a marked reduction in frequency of appearance of the variegated class of kernels and an equivalent increase in 

the number of colored kernels. In the pale class, pigment intensity was greater when Pr was present than when the kernels were homogeneous for Pr. However, the Pr-carrying kernels in the pale class could be distinguished readily from those that exhibited the full or near full A1 type pigmentation. This is in contrast to the pale, Pr class produced by state 

A1 where difficulties were encountered in making this distinction, as described in the previous section.

From the basis of the ratios given in table 10, the unselected constitution of those plants with respect to Spm is given in the last column. Tests conducted with different parts of a single plant agreed with another that which produced the ear on the main stock of plant A-5. In this respect, it is concluded that plants A-2, A-3, A-4, A-6 and A-8 had one Spm, whereas plant A-5 had two Spm elements located at a different positions in the chromosome complement.
Nearly all of the variegated kernels on ears produced by use of pollen of 1 Spm carrying plants on silks of plants homozygous for $a_1$ (C, table 10) had, in addition to dots of $A_1$-type pigment, areas of various sizes exhibiting the same grade of pale pigment that is present in the pale class of kernels. In other words, these kernels were similar in phenotype to the kernel from which each plant arose. Some kernels of this type likewise appeared among the variegated class on the ear produced from use of pollen kernels exhibiting dots $A_1$ in a silver background from plant A-5. Among the variegated class of kernels produced by the reciprocal cross, B, table 10, those exhibiting pale areas that were of various sizes were rare. Pale areas were present in some kernels but they were small in size and few in number. It was learned in the previous section that such pale areas could be expected to appear if, for some reason, the Spm element (s) in them is lost from an endosperm cell during development of the kernel. If one Spm element is present, loss of it from some cells at various stages in development would result in the appearance of pale areas of various sizes. If two Spm elements were present, simultaneous loss of each or successive losses of them would be required in order that a pale area would appear. Because the female parent contribute two identical nuclei to the endosperm and the male parent only one, the initial Spm number in the endosperms of variegated kernels,
entered in B of table 10, would be at least two whereas the initial Spm number in the majority of those produced by the cross of plants of A-2, A-4, and A-8 in C of this table would be but one. It was obvious from observations of kernel types derived from all crosses entered in table 10 that a relation existed between the number of Spm elements in a kernel and the frequency of appearance, the size range of pale areas in it. The Spm elements was being subjected to some type of modification but the nature of this was not discovered until later.

From the same ear that produced the kernels from which the plants in 6081A were derived, another variegated kernel was selected. It exhibited only the deeply-pigmented dots. No pale areas were present. Plant 6081B was grown from it. The types of kernels produced by self-pollination and reciprocal crosses with plants homozygous for \(a_1\), are entered in table 11. The ratio of kernel types on these ears suggest that this plant had more than two Spm elements in it and these ratios conformed with those obtained from the parent \(a_1^{m-1}\) plant, 5719A-2 (A, B, and C of table 2). It would appear, then, that selection of variegated kernels exhibiting pale areas of various sizes in addition to the dots with \(A_1\) type pigment in them, is an effective method for selecting those kernels on an ear that have only one or occasionally 2 Spm elements in them.
A test conducted with plants derived from variegated kernels on the self-
pollinated ear of plant 5719A-2 did not negate this.

From the self-pollinated ear of plant 5719A-2, variegated kernels
having only dots of A₁ were sown under culture number 6080C. There
was one variegated kernel on this ear that exhibited a number of pale areas
in addition to the A₁ dots. The plant grown from it was given culture
number 6080B. Reciprocal crosses of this plant to plants homozygous for
a₁ produced the kernel types entered in table 12. This plant proved to be
homozygous for the 5719A-2 state of a₁₄. From the ratio of kernel
types on these ears, it may be concluded that plant 6080B had only one Spm
element in it. In contrast, among the seven plant in C of this culture
that also were homozygous for this state of a₁₄, only one had a single
Spm element in it, and the ratio of kernel types appearing on ears produced
by self-pollination of these plants and by reciprocal crosses of them with
plants homozygous for a₁, given in A to C of table 13, indicate this.

Tests conducted with some of the progeny derived from plant 5719A-1
were discussed in previous sections. It was obvious from them that plant
5719A-1 had more than one Spm element in its nuclei. Kernels derived
from self-pollinated ears of this plant were grown under culture numbers
10466A, 10466B, and 10466C. On these ears, there was a small number of variegated kernels

that exhibited some pale areas in addition to the small spots, with one type pigment in them. Three plants were derived from kernels of this type. Tests of them indicated that in two of these three plants one Spm element was present, and in the third, two Spm elements were present, as the second, third, and fourth plants in table 15. Data in table 14 indicate that the variegated kernels on ears entered in C of this table were characterized by the appearance of pale areas in addition to the $A_1$ spots, whereas these pale areas were rarely exhibited among the variegated class of kernels in B of this table.

Variegated kernels derived from the self-pollinated ear of plant 5718 (A, table 2) were sown under culture number 6045. Although 16 kernels were sown, only 4 plants derived from them survived to maturity. Two of them were self-pollinated and crossed to plants homozygous for $a_1^m$. The type of kernels on the resulting ears are shown in A and B of table 15. The pattern of variegation produced by the 5718 state of $a_1^m$ is very sharply expressed. There are small dots of the $A_1$ phenotype in a colorless background, as shown in photo 00. In the absence of Spm, this state gives rise to kernels that are only very faintly pigmented. Difficulty was encountered in detecting this pigment in some kernels on the ears entered in table 15, that should be carrying this state of $a_1^m$ but no Spm. Unless this pigment was undoubtedly present, the kernel was placed in the colorless ground.