Cytoplasmic inheritance of albinism in a striped mutant of titan barley
by Saidollah Kazemi

A thesis submitted to the Graduate Faculty in partial fulfillment of the requirements for the degree of
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Abstract:
A spontaneous striped mutant of Titan barley, which upon selfing yields striped and albino seedlings with almost equal frequency, was investigated to determine the genetic basis of its variegation.

Translocation stocks were used to investigate the location of gene for striping. Physical and chemical characteristics of this mutant were observed and compared to the normal Titan barley.

Our studies in chambers with different temperatures revealed that the gene for striping requires a threshold, that is, plants raised at colder temperatures indicated a higher rate of striping than seedlings grown at warmer temperatures. Seeds of striped mutant raised under different temperatures yielded different ratio of albino seedlings. Seeds of striped mutant, raised in several different locations in Montana, also seemed to yield different ratios of albino seedlings.

Yield trials of normal Titan conducted in nine different locations, compared to the striped mutant, showed that in spite of doubling the seeding rate for the mutant type to allow for the loss of albinos from the stand, it outyielded the mutant type significantly.

Trials conducted at different stations in Montana indicated that this mutant had higher protein level and agtron readings than the normal Titan barley.

By separating the large seeds from wrinkled and smaller ones, it was observed that the shriveled seeds almost always gave rise to albino seedlings. The albino seedlings emerging from these seeds often died before, they reached the surface of the soil.

Reciprocal crosses conducted showed that when the striped mutant was the maternal parent and translocation stocks as the pollen donors, the F1 yielded striped and,albino seedlings; and its reciprocal cross yielded, only green seedlings, thus indicating a definite cytoplasmic inheritance for the albinism. These crosses also indicated that the gene for striping is recessive to its dominate allele and upon homozygosity causes striping of the leaves.

Linkage studies with translocation tester stocks showed that the mutant gene is possibly linked with translocation points T3-5b and independent of Tl-7a, T2-4a, T2-6a, and T3-7a, thus indicating that maybe the gene is located on chromosome number five or three.

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ABSTRACT

A spontaneous striped mutant of Titan barley, which upon selfing yields striped and albino seedlings with almost equal frequency, was investigated to determine the genetic basis of its variegation.

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Our studies in chambers with different temperatures revealed that the gene for striping requires a threshold, that is, plants raised at colder temperatures indicated a higher rate of striping than seedlings grown at warmer temperatures. Seeds of striped mutant raised under different temperatures yielded different ratio of albino seedlings. Seeds of striped mutant, raised in several different locations in Montana, also seemed to yield different ratios of albino seedlings.

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Trials conducted at different stations in Montana indicated that this mutant had higher protein level and agtron readings than the normal Titan barley.

By separating the large seeds from wrinkled and smaller ones, it was observed that the shriveled seeds almost always gave rise to albino seedlings. The albino seedlings emerging from these seeds often died before they reached the surface of the soil.

Reciprocal crosses conducted showed that when the striped mutant was the maternal parent and translocation stocks as the pollen donors, the F_1 yielded striped and albino seedlings; and its reciprocal cross yielded only green seedlings, thus indicating a definite cytoplasmic inheritance for the albinism. These crosses also indicated that the gene for striping is recessive to its dominate allele and upon homozygosity causes striping of the leaves.

Linkage studies with translocation tester stocks showed that the mutant gene is possibly linked with translocation points T3-5b and independent of T1-7a, T2-4a, T2-6a, and T3-7a, thus indicating that maybe the gene is located on chromosome number five or three.

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INTRODUCTION

A gene mutation may be defined as a change of one base pair in the DNA chain allowing a detectable and permanent alteration of the phenotype. Distinguishing point mutations from larger changes involving segments of chromosomes containing many genes may be difficult. Gene mutations could arise spontaneously or they could be induced artificially by chemical and/or physical means such as temperature.

Here we have undertaken to investigate a striped mutant barley which arose spontaneously in the variety Titan, and upon selfing, yields striped and albino seedlings.

Various experiments were conducted to describe its effect on phenotype. By marking the lemma and palea of the seeds which were green or white, their future development into albino and striped seedlings were studied.

After hand separation of seeds of the striped mutant into "small" and "large" seeds, they were planted in the greenhouse and their segregation into striped and albino seedlings was studied.

Seeds of the striped mutant raised at various locations were planted in the greenhouse and the percentage of albino seedlings was observed.

To investigate and compare the agronomic performance of this mutant to normal Titan, yield trials of both types of barley were conducted at several locations in Montana.

The malting quality of the mutant was determined and compared to that of normal Titan.

To estimate the effect of environment on the mutant and the amount of striping, seeds of the striped mutant were raised in chambers with varying

temperatures, and the albino areas of the leaves of striped plants were measured.

Seeds of the mutant raised under various temperatures were planted in the greenhouse to investigate the effect of temperature on the seed and relating to their development into striped or albino seedlings.

Reciprocal crosses of the striped mutant and normal Titan were made to detect the possibility of extra-nuclear inheritance for the albino character.

Translocation stock were used to investigate the location of the gene ws ws (striping gene) and msl msl (male sterility).

Linkage intensities between the gene green vs. striped mutant (Ws ws), male fertile vs. male sterile (Msl msl), male fertile vs. male sterile (Ms8 ms8), fertile vs. sterile laterals (Ii), long vs. short awned glumes (Ee), rough vs. smooth awns (Rr), and two row vs. six row (Vv) were investigated.

REVIEW OF LITERATURE

A gene mutation may be defined as a change of one base pair in the DNA chain allowing a detectable and permanent alteration of the phenotype. In practice, it may be difficult to distinquish point mutations from grosser changes involving segments of chromosomes containing more than one gene. Such gene mutations arise spontaneously; or they may be artificially induced by heat or chemical treatments, ageing, or various kinds of ionizing radiations. Point mutations, in the strict sense, should be transmitted in approximately normal, theoratical ratios. On the other hand, grosser changes involving chromosomal aberrations should modify the transmission of effected chromosomes, resulting in abnormal ratios of genes associated with them.

Aberrant Albino Ratios

C. Moh, L. Smith, Nilan (10, 11, 12, 13, 23) did a considerable amount of work involving atomic bomb, x-ray-induced, and spontaneous seedling mutants in barley and durum wheat. They recovered various percentages of mutant seedlings, and differences between mutant types from the bomb, x-ray, and spontaneous source were not large enough to be of any apparent biological importance. There were also no clear differences in the transmission of mutants in X_3 and X_4 generations.

More than 20% of the mutants, tested in X_3 and X_4, segregated in ratios significantly different from the theoratical 3:1. Most of the mutants segregating in aberrant ratios were associated with a low transmission of the mutated gene.

Germination in most of the progeny tests was over 90%. Variations in germination did not seem to be associated with the percentage of recessive segregates. Apparently the aberrant segregations were not the result of differential seed germination per se.

They concluded that chromosomal aberrations (possibly deficiencies) associated with a considerable portion of these mutations, or pleiotropic effects of mutant genes, best accounted for the fact that over 20% of the mutants tested segregated in ratios that deviated significantly from the theoretical 3:1. The results could readily be accounted for, if it were assumed that a considerable portion of the mutations were in fact deficiencies rather than gene or "point" mutations.

Maternal Inheritance

At the present time, it is commonly believed that the plastid is an independent self-reproducing entity capable of undergoing mutation. The basic unit of the plastid has been called plastogene and may be, at certain stages, no more than macromolecular size. The plastogene belongs with those protoplasmic elements, gene, plasmagene, and virus particle--which are able to duplicate themselves. That genes, plastids, and viruses all contain or consist of nucleoproteins is of more than passing interest in this connection.

A large number of cases of non-mendelian heredity of plastid differences (variegation or striping), where the transmission is not influenced by a nuclear gene, have been described. The most common of these is that in which the plastid variegation is inherited through the mother only.

The first example of this kind was described by Correns, as cited by Rhoades (20). While he was working with green-white variegated plants of Mirabilis jalapa, the following breeding behavior was found: (1) Flowers on green sectors gave only green offspring, irrespective of the constitution of the pollen parent. (2) Flowers from white branches produced only white seedlings, no matter what kind of pollen was applied to the stigmas, while (3) flowers from variegated branches yielded a mixed progeny of green, white and variegated plants in widely varying degrees.

The simplest and most widely accepted view is that the variegated plants possess two distinct kinds of plastids (green and white). It is assumed that the white type arose through mutation and that the two types became separated by somatic segregation during cell division. Baur, Gregory, Winge, Wettstein, Imai, as cited by Rhoades (20), and many others hold to this explanation. The hypothesis of somatic segregation of mixed plastids demands a genetic autonomy and independence of the plastid types. The breeding behavior is seemingly in harmony with the hypothesis, but cytological observations yield discordant results. Gregory, as cited by Rhoades (20), found two types of plastids in Primula sinesis albomocultata. The mature green portions of variegated plants had only normal plastids and the chlorotic areas had only smaller, nearly colorless plastids, while both types were observed in embryonic cells. Similar observations on different plant species have been reported by other investigators. Here, then, appears to be the cytological confirmation needed to complement the genetic data.

Correns, as cited by Rhoades (20), believed that a diseased cytoplasm was responsible for the variegation of albomoculata plants; but later adapted the view that the cytoplasm in embryonic cells of variegated plants exists in an indifferent labile condition, which changes fortuitiously into a normal state that permits plastid development or into a diseased condition that prevents normal plastid development.

East, as cited by Rhoades (20), thought it probable that an active parasitic agent present in the cytoplasm is responsible for cytoplasmically inherited chlorophyll variegation.

Rhoades (19) agrees with Correns that some cytoplasmic condition other than the plastids furnishes the correct explanation for the type of inheritance found in "status albomoculatus".

Rhoades (19) proposes that cells with a critical percentage of abnormal plasmagenes would contain poorly developed plastids, while those cells with higher proportions of normal plasmagenes would have more normally developed plastids.

Variegated common wheat was reported for the first time by W. K. Pao and H. W. Li (16). Selfing the variegated plants yielded green, variegated and pure yellow individuals in the next generation.

Progeny from selfed variegated plants varied from year to year and from family to family. Variegated plants had yellow stripes on glumes, lemmas, paleas and even anthers. By marking the glumes, they followed the development of the seeds within their glumes into seedlings in the next generation. They found that the seed from glumes that were yellow gave

rise to yellow seedlings, green glumes gave rise to green seedlings, and seed from variegated glumes developed into variegated plants.

They made crosses using green plants as the female and variegated or yellow plants as males. The F_1 seedlings were all green and no segregation of either pure yellow or variegated took place in F_2. When variegated plants were used the female parent, the F_1 seedlings were either all pure yellow or green or variegated, depending on the type of sector on the mother plant where the seeds developed. The segregation of F_2 and F_3 from the variegated F_1 plants was like that of the original stocks.

They also used a variegated plant as the female parent in a cross with T. turgidum. The F_1's were pentaploid hybrids, each with 35 chromosomes. Three of the four F_1 plants were yellow-striped, and the other was normal green. The F_2 and F_3 of the variegated ones segregated into green, variegated and yellow as usual. It appears, therefore, that the inheritance of the yellow-striped character is not altered by the irregularity of the meiotic division of these pentaploid hybrids.

In these cases, yellow-striped character of wheat is transmitted exclusively through female gametes (maternally inherited).

One example of cytoplasmically inherited chlorophyll variegation in maize have been reported by E. G. Anderson (2).

Cytoplasmic-genic Interaction for Inheritance of Plastid Abnormalities

There are hundreds of examples of gene-controlled plastid characters. In maize, for example, there are more than a hundred genes that produce a wide range of mutant phenotypes such as white, yellows, stripes, etc. In

many of these cases, it is likely that gene effect has been merely the production of a physiological condition unfavorable for normal plastid development; it is also possible that mutations of the plastogenes have been induced. That mutation of the plastogene can occur through genic action has been reported by Sô for barley, as cited by Imai (5), Imai (6, 8) for barley, Pai (15) for rice, Rhoades (18) for maize, and Cleland (4) for Oenothera.

There is no reasonable doubt that gene induced cytoplasmic mutations occur, that the cytoplasm, or, more likely, some particulate part thereof can be permanently modified by specific genes (21, 22). Two significant studies were published by Spiegelman, Lindegren and Lindegren (20, 27). They found an adaptive enzyme in yeast that was gene-initiated but self-duplicating in the presence of substrate, even though the activating gene was absent. Sonneborn (24, 25, 26) found that the K gene in Paramecium aurelia could form a cytoplasmic substance, Kappa, when Kappa was already present in the cytoplasm, but could not initiate the formation of Kappa.

Early work on variegated (striped) barley was done by Y. Imai (5). He studied variegated barley and also contributed to this field by translating and discussing the work of Sô and other Japanese scientists who were studying variegated barley.

Among the specimens of barley grown at Tokyo Imperial University, Sô came across a variegated variety of barley which he named "Okina" (old man). The name refers to the white awns of barley which resembled the white hair of an old man. The progeny of this variety segregated into white and variegated.

The variegated characteristic segregated as a Mendelian recessive to normal green. Sô, as cited by Imai (5), studied the genetic behavior of this variegated variety and concluded:

1. The albinism is transmitted only maternally, the pollen having no immediate connection with it.
2. Variegation behaves as a recessive to green.
3. In F_2 and later generations, the segregation of variegation occurs in monohybrid ratios.
4. In crossing experiments, the pollen from flowers with pure white glumes is not different in its effect from that of green ones of the variegated ear (head).

Selfing the variegated variety, he found about 20% albinos and 80% variegated. In crosses with normal green as the female and variegated as male, the F_1 were all green and the F_2 segregated 3 green:1 striped. When pollen from normal green was transferred to the variegated female, the F_1 gave 4 green:1 albino (proof of maternal inheritance).

Sô proposed that gene w in heterozygous form is responsible for the partial mutation of plastids and variegation on the plants; and when in homozygous form, it causes complete mutation of the plastids and, as a result, we recover albino seedlings.

Imai (7) studied the effect of environment on the rate of plastid mutations in Hordeum vulgare. In his experiments, he tested the effect of sowing time of this variegated barley. To do this, he planted seeds of variegated barley at 10 day intervals (from September 10 until November 9). The later the seeding, the lower the percentage of albino seedlings in the segregating progenies. The percentage varied from 11.3% to 3%.

He pointed out that although differences in sowing time may accompany many environmental factors which influence the mutability of the plastids, the most important of these factors seems to be temperature.

Similar work was done by Eyster, as cited by Imai (7), when he showed the effect of environment on variegation patterns in maize pericarp.

Imai (6) suggested that self-reproducing plastids contain plastogenes. Mutation of plastogenes may be induced by certain nuclear genes, and automutation independent of nuclear genes may also occur. When only a portion of the plastids of meristematic cells mutate, e. g., embryo, segregation of plastids may occur at successive cell divisions resulting in the characteristic green and white sectors. Plastid mutation may occur commonly only at certain stages of the life cycle, e. g., in germ cells or cells of young embryos.

Arnason, et al., (3) working with a strain of variegated barley, that originated in a Saskatchewan field, found that it produced on selfing, progeny of which approximately 90% were albinos, the remainder striped or variegated with rare full green exceptions. Crosses of variegated female X green male produced 7 albino, 4 striped, and 11 green F_1 plants. The reciprocal cross yielded 1 striped and 4 green F_1 plants. The F_2 segregation of both crosses, approximated three green to one of all others (albino and variegated). The pecularities of inheritance was explained on the basis of a combination of gene and (maternal) plastid inheritance. On this interpretation, the plastids present in the egg affect the color of the seedling that develop from it. When green plastids or proplastids are present in the egg, many of them, but not necessarily all, are induced

to mutate if the white (w) gene is homozygous, but fewer if w gene is heterozygous. If white plastids only are present in the egg, it is probable that the seedling will be an albino regardless of gene content.

Among the chlorophyll characters in maize, which are genically controlled, is that of iojap. Maize plants homozygous for the recessive gene iojap (ij) exhibit a chlorophyll striping or variegation. This was shown by Jenkins, as cited by Rhoades (18). Further work by Rhoades (18, 20) confirmed Jenkins findings; he found that the recessive iojap (ij) gene, located on chromosome 7 in maize, produces green and white striped plants when homozygous. When ears of iojap plants are crossed with pollen from unrelated green plants, the offspring may consist only of normal green plants or may consist of green, white and green-white striped plants in widely varying ratios. Occasionally an F_1 progeny is comprised only of white offspring, but this is relatively infrequent. The reciprocal cross of green females by iojap males yields only green plants.

When the F_1 striped plants, which are all heterozygous for the dominant allele of iojap, were pollinated by green males, the progenies consisted either of all white, or all green, or of a mixture of green, white, and occasionally a few striped individuals. In the all white progenies, half of the plants were homozygous for the normal allele of iojap; yet all plastids were small and colorless. It was concluded that the iojap allele includes irreversible plastid mutations and that these mutant plastids retain their changed characteristics in outcrossed generations even when the inducing gene has been wholly replaced by the normal allele.

A definite line of evidence for the genetic continuity of plastids comes from quite a different type of observation; namely, the exhaustive survey of species crosses in Oenothera. These were started by O. Renner and continued by W. Stubbe, T. Stenson and R. E. Cleland (4), and are cited by Jinks (9). They concluded that the reaction between the plastids and the genome is that both components are equally permanent and autonomous. The function of the plastids was upset in a foreign genome, but the plastids themselves remained uneffected when recombined with their own genome.

The plastids of O. muricata and O. hookeri are demonstrably different in their reaction to an identical genome, and their differences persist undiminished by their association with a foreign genome. This implies permanent, heritable differences between the plastids of these two species.

The plastid differences could have arisen only during or after the separation of the species, which now constitutes this genus. It cannot be doubted, therefore, that plastids may change by mutation or that the changed forms are capable of self-perpetuation.

MATERIAL AND METHODS

The mutant studied here was found in a field of Titan barley. Its appearance differs from the normal Titan in having stripes of white or light green and green sectors on the leaves at early stages of growth; and during the heading stage, this variegation in color appears on glumes, lemma, palea, awns, and even on anthers. Because it gives rise to some white awns, it has been called "Grandma Titan".

Studies concerning the yield of the striped mutant in comparison to normal Titan were conducted at nine different locations in the state of Montana, under irrigated and dryland conditions. Seeding rates of the white striped mutant were doubled to allow for the loss of albinos from the stand. Five replications of 4 row plots were grown at each location. Seeds of this mutant type raised since 1954 at different locations and in different years at the same location were available for study.

In our studies concerning the effect of the temperature on the amount of striping and its future segregation into albino or striped plants, we used three growth chambers with controlled temperatures of 2-10, 10-16, and 21-27 degrees centigrade.

Measurements of white and green sectors of the leaves were done by using a calibrated slide and eyepiece on a dissecting light microscope. Measurements of striping were made when the plants were about three weeks old, and the second leaves from the bottom of the plants were selected for this study. To compare the amount of striping of the first and fifth leaves of the plants raised from striped mutant seeds grown under different temperatures, we waited until the heading stage before the measurement of stripes.

Chemical and physical data comparing the striped mutant to normal Titan samples grown at different locations were recorded in 1960. Student's pairing method was used to show if there were any significant differences in these data between the mutant and the normal Titan.

Malting evaluations were done at the USDA Malt Laboratory at Madison, Wisconsin through the courtesy of Dr. A. D. Dickson.

To study the relationship of the lemma color at heading stage to the production of striped and albino seedlings, two different colors of India ink were used. Red ink was applied to the lemma which displayed light or white color at heading stage, while black India ink was applied to the predominately green lemmas. After maturity, these marked seeds were raised in the greenhouse and the seedling color recorded.

When the author started his work in this field September, 1965, the F_1 population of the mutant Titan by translocation tester stocks was growing in the greenhouse.

The F_2 generation was space planted in the spring of 1966. Planting was done by means of a hand drill that planted 20 seeds within a 3 meter row with 30 Centimeter between rows. Striped mutant plants in the F_2 generation were marked with tags. Identification of striped plants was best achieved during the heading stage, and presence of white awns and variegated heads were definite signs of striping.

Male sterile, semi-sterile and fertiles in the F_2 generation were recorded after maturity. To be sure of correct reading of these plants during the harvest period, each individual plant was pulled and read for its fertility. Sterile plants had few or no seeds on its heads, while

semi-sterile ones had all their seeds except a few central and laterals were missing; fertile heads had all seeds present.

Segregation of F_3 hills for striped or green and fertile or male sterile was checked at heading stage. To make sure of our readings on male sterile individuals, we examined their anthers which were shriveled as compared to anthers from normal heads. Phenotypic features, like opened lemmas and paleas, were quite easy and rather reliable in predicting sterility. Fertile and semi-steriles had to be read after the heads matured.

Our F_2 population in the summer of 1966 contained crosses of translocation stocks, T1-7a, T2-4a, T2-6a, T3-5b, and T3-7a in male sterile form by striped mutant. Seven reciprocal crosses of striped mutant by translocation stocks were made during the summer and their F_1 generations were raised in the greenhouse during the winter period 1966-67. F_2 for these crosses along with the F_3 population of the other translocation stocks crossed with the striped mutant were planted in the spring 1967.

Linkage intensities between the genes stripes mutant (ws ws), male fertile vs. male sterile (Ms1 ms1), male fertile vs. male sterile (Ms8 ms8), fertile vs. sterile laterals (Ii), long vs. short awned glumes (Ee), rough vs. smooth awns (Rr) and two row vs. six row (Vv) were investigated.

Our F_2 population consisted of around 2700 plants, while the number of plant hills for our F_3 generation was reduced to about 850 hills. The studies at the Plant and Soil Science Department of Agricultural Experiment Station at Bozeman are under irrigated conditions. Hills were planted by making a hole in the field .6 meter from the next hill and by placing a

single head in each hole.

To observe and record the existence of albino seedlings of the segregating hills, they were checked and rechecked at early stages of growth to make sure that the existence of albino seedlings were recorded before the lack of chlorophyll in these seedlings caused them to perish.

Studies of cytoplasmic inheritance were done on F_3 generation by transferring pollen from normal Titan or other varieties of barley to male sterile heads of the striped mutant. The reciprocal cross for this experiment was made in the summer of 1965. F_2 generations for this cross were studied in the summer of 1966 and F_3 generations for this cross were investigated in the summer of 1967. In our crosses of the striped mutant by normal Titan during the summer of 1967, 1000 PPM Gibrellic acid was used to soak the seeds and reduce the dormancy period of the newly formed seeds, thus inducing faster germination of these seeds.

RESULTS AND DISCUSSION

Relation of Striping to Percentage Albinos

To observe the relationship of lemma color at the heading stage to the production of striped or albino seedlings, India ink was used to mark the two distinctly colored lemmas--those being white or predominately white and those that were green or predominately so. These seeds were later grown in the greenhouse and the record of their development into striped or albino seedlings are summarized in Table I.

As indicated by the percentage of albino seedlings, there is a definite relation of lemma color to future development of the seed into striped and albino seedlings. The highly significant interaction X^2 supports this observation.

It is concluded that seeds formed within white lemmas and paleas which possess white or mutated plastids will give rise to a higher frequency of albino seedlings than seeds formed within green lemmas and paleas.

The relative number of albino seedlings in the next generation depends on the amount of striping on the parental plant. The greater the amount of striping in the mutant plant, the more albino seedlings in the next generation. Both high and low striped plants were marked in the field and their selfed seeds were collected. After germination, it was learned that 42 out of 76 of the seedlings arising from highly striped plants were albinos, while low striped plants yielded only 1 albino seedling to 65 striped ones.

Relation of Seed Size to Percentage Albinos

To compare large seeds and smaller seeds from a stock of striped

Table I. Relationship of lemma color on striped Titan plants at heading stage to the production of striped and albino seedlings after selfing.

Florets Marked For:	No. of Seeds		Seedling Color		Percent Albino
	Planted	Emerged	Striped	Albino	
Green	86	83	68	15	18.1
White	58	45	14	31	68.8

Interaction X^2 for seedling color = 31.75, $P<.01$

mutant, with respect to their development into striped and albino seedlings, seeds were hand selected into very large and very small seeds and planted in the greenhouse. The percentage of albinos resulting from their germination is given in Table II.

Calculated interaction X^2 indicates that there is a definite pattern of development of smaller seeds into albino seedlings, while large seeds develop into striped seedlings.

Effect of Seed Source on Percentage of Albinos in Population

Seeds of striped mutant raised at different locations and in different years were collected and germinated in the greenhouse. Their segregation into striped and albino seedlings are reported in Table III.

Interaction X^2 for seedling color was applied and are listed in Table III. In 9 cases out of 12, it indicated an acceptable P level; but a calculated heterogeneity X^2 indicated that these seeds from different sources are hetergenous for their segregation into striped and albino seedlings. In one instance, more than expected; and in the other, less than the expected number of albino seedlings were observed. This difference in their segregation pattern was later attributed to the environment under which the seed was grown.

Agronomic Performance of the Mutant

To study the chemical and physical characteristics of the striped mutant, seeds of this mutant along with normal Titan were raised at several different locations throughout the state of Montana. Table IV gives percent protein, kernal weight, percent plumpness and average agtron readings

Table II. Comparison of large seeds vs. smaller seeds from striped plants and their development into striped and albino seedlings.

Seed Size	No. of Seeds		Seedling Color		Percent Albino
	Planted	Emerged	Striped	Albino	
Very large	108	87	72	15	17.2
Very small	109	69	17	52	75.3

Interaction X^2 for seedling color = 53.17, $P<.01$

Table III. Seeds of striped mutant raised at different locations, conditions and their development into striped and albino seedlings.

Year	Location	Condition	No. of Seeds Planted	No. of Seeds Emerged	No. of Seedlings Striped	No. of Seedlings Albino	Percent of Albinos in Population Emerging	Interaction X^2	P Level
1954	Bozeman	Irrigated	400	163	81	80	50.3	2.67	.50-.10
1955	Bozeman	Irrigated	400	340	163	177	52.1	10.48	P<.01
1956	Bozeman	Irrigated	400	314	173	141	44.9	.42	.90-.50
1958	Bozeman	Irrigated	400	339	198	141	41.6	.32	.90-.50
1959	Bozeman	Irrigated	400	292	147	145	49.6	4.76	.05-.01
1965	Bozeman	Irrigated	200	105	66	39	37.1	1.68	.50-.10
1966	Bozeman	Irrigated	400	339	188	151	44.5	.18	.70-.50
1960	Creston	Dryland	400	350	196	154	44.0	.061	.90-.80
1960	Creston	Irrigated	400	315	312	102	32.3	15.4	P<.01
1960	Havre	Irrigated	400	334	193	141	42.2	.17	.70-.50
1960	Sidney	Irrigated	400	359	217	142	39.5	2.08	.20-.10
1960	Sidney	Dryland	400	317	186	131	41.3	.51	.50-.30
All Samples			4600	3567	2021	1546	43.3	.222	.90-.50

Sum of X^2's = 39.04 Heterogeneity X^2 = 38.818, P<.01

Table IV. Chemical and physical data on sample of white striped Titan mutant as compared to Titan grown at several locations in 1960.

Location	Condition	Percent Protein		100 Kernal Weight Grams		Percent Plumpness		Avg. Agtron Readings	
		Normal	Striped	Normal	Striped	Normal	Striped	Normal	Striped
Bozeman	Irrigated	15.3	18.1	3.15	3.42	60.1	57.2	59	64
Havre	Irrigated	15.0	15.9	2.93	2.88	13.9	12.1	71	71
Sidney	Dryland	16.0	17.8	2.69	2.70	12.2	20.0	53	65
Sidney	Dryland	11.0	14.3	2.62	3.77	68.4	57.8	53	62
Creston	Dryland	12.3	13.9	2.75	2.67	26.8	24.9	57	62
Creston	Irrigated	15.8	17.8	3.69	3.50	66.2	56.8	41	49
Moccasin	Dryland	16.7	17.5	2.19	2.22	0.7	1.6	79	77
Huntley	Dryland	18.0	17.8	2.32	2.98	0.7	1.6	44	60
Huntley	Irrigated	16.5	19.4	2.93	2.50	20.3	32.8	60	62
Average		15.2	16.9*	2.81	2.96	29.9	29.4	57.44	63.44*

* Significantly higher at P = .05

for the Titan mutant and normal Titan.

Student's pairing method of calculation of t was applied and indicated significantly higher values for the mutant type for percent protein content and average agtron readings. The agtron reading indicates the amount of light reflection from the harvested seed.

Yield trials of the striped mutant in comparison to normal Titan were conducted at several different locations under dry or irrigated conditions and are listed in Table V. Seeding rate of the mutant was doubled to allow for the loss of albinos from the stand. The mutant type was lower in yield than the normal Titan in all trials, and is significantly so in most of the trials conducted. A complex analysis of variance of striped mutant vs. normal Titan is given in Table Va. It indicates that there is a consistent significant difference in the yield of striped mutant vs. normal Titan at all locations.

Observations made here, concerning the yielding ability of the mutant, indicate that the mutant gene not only is lethal in producing albino seedlings; but also that the striping limits the growth of plants, thus reducing the yielding capacity of the mutant type.

Table VI gives a comparison of malting quality of mutant to normal Titan samples from several locations. The observed differences are consistent with the observed differences in protein content. It is proposed that due to higher protein level in its seeds, the striped mutant type would be inferior in its malting quality.

Table V. Effect of environment on the comparative yield of white striped and normal Titan at different locations.

Location	Condition	Mean Yield in gm./16 sq. ft. Green	Striped	Mean Square Green vs. Striped Titan with 1 Degree Freedom
Huntley	Dryland	112	66	5,428*
Huntley	Irrigated	402	365	3,460
Creston	Dryland	130	94	3,240*
Creston	Irrigated	352	289	10,049*
Havre	Dryland	175	129	5,198*
Sidney	Dryland	246	189	7,562**
Sidney	Irrigated	651	529	37,577**
Moccasin	Dryland	154	147	145
Bozeman	Irrigated	485	287	99,813**
All Locations		300.8	232.6	103,972*

* = significant at 5% level.

** = significant at 1% level.

Table Va. Complex analysis of variance of yield of striped mutant Titan and normal Titan grown at different locations.

Source of Variations	Degrees of Freedom	Mean Square
Replications within Locations	36	1,949
Location	8	274,926**
Striped Mutant vs. Normal Titan	1	103,972**
Striped Mutant vs. Normal Titan X Location	8	7,312

** = significant at 1% level.

Table VI. Malting quality of striped mutant and normal Titan barley.

Characterization	Striped Mutant	Normal Titan
Barley N, %	2.85	2.64
Barley kernel wt., mg.	29.0	30.5
Plump barley, on 6/64 sieve, %	32.8	59.8
Barley color, agtron	54.0	45.0
Malt extract, dry, %	71.1	71.8
Wort N, %	.895	.825
Wort N/malt N, %	30.3	31.5
Diastatic power, ^{o}L	265	243
Beta amylase, maltose equiv.	821	776
Alpha amylase, 20^{0} dex. units	72.4	60.4
Ratio beta/alpha amylase	11.3	12.8

Effect of Temperature on Striping

Striped mutant plants grown under different temperatures express different degrees of variegation. Those raised at colder temperatures, (2-10° C.), display more striping than the ones grown at warmer temperatures, (10-16 and 21-27° C.). Five leaves from five different plants were selected for each measurement. These measurements and the analyses of variance are given in Table VII. The significant F value confirms the hypothesis that at colder temperatures, the amount of striping is higher than at warmer temperatures. A test for least significant difference between the amount of variegation at 10-16 and 21-27° C. indicated a 3.56 value which is larger than the difference between the mean percentage of striping at these temperatures, thus indicating that the difference between the amount of striping at 10-16 or 21-27° C. is nonsignificant.

In another study, seeds of striped mutant which had been raised under different temperatures were planted in the greenhouse and the extent of leaf striping and percentage albino seedlings were determined.

The calculated F value for the first leaf indicates a significant difference in the amount of striping for seeds raised under different temperatures, while the differences in the amount of striping of the fifth leaf is nonsignificant (Table VIII). The colder the temperature, the greater the amount of striping on the first leaf. The difference in the amount of striping of the first and fifth leaf is attributed to the fact that the first leaf primordin was already formed in the embryo, while the seed was developing under the specified temperature. When the first leaf arises, it expresses its striping in accordance with the temperature at which the

Table VII. Effect of germination temperature on the amount of variegation on the second leaf of striped mutant.

Temperature, Degrees Centigrade	Mean Percentage of Leaf Width that is Albino
2-10	38.3
10-16	17.9
21-27	15.5

Analysis of variance for percentage of albino areas of leaves raised under 3 different temperatures.

Source of Variation	Degrees of Freedom	Sum of Square	Mean Square	F Value
TOTAL	14	1960.1	140	
Treatment	2	1576.4	788.2	24.7*
Within Treatment	12	383.7	31.9	

* = Significant at 1% level.

Table VIII. Percentage albino area of the first and fifth leaves of striped mutant plants grown from seeds raised under different temperatures.

Temperature, Degrees Centigrade	Mean Percentage of Leaf Width that is Albino	
	First Leaves	Fifth Leaves
2-10	60.7	41.4
10-16	40.1	37.5
21-27	33	35.1

Analysis of Variance for Percentage Area for:

Source of Variation	First Leaves		Fifth Leaves	
	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square
TOTAL	13	273.2	14	32.04
Treatment	2	891.5*	2	55.6 N.S
Within Treatment	11	160.9	12	28.12

* = Significant at 5% level.

N.S = Nonsignificant

seed was raised; and since the growth of the plumule of the first leaf of the embryo was under the same temperature at which the seed was raised, it displays high striping when the parental seed was formed under cold temperature, and low striping when the seed was formed under warmer temperature.

Seeds of the striped mutant formed under different temperatures were planted in the greenhouse and percentage albino seedlings counted and are listed in Table IX. In the previous experiment concerning lemma color and its future development into striped or albino seedlings, we indicated that highly striped plants give rise to a higher ratio of albino seedlings, while the results presented in Table IX indicate just the opposite. The low percentage of albino seedlings resulting from seeds matured at low temperatures (highly striped plants) could be explained on the basis of the observed low fertility of plants grown at cold temperatures. It could be assumed that florets with white lemmas which would produce seeds that would develop into albino seedlings did not survive the temperature stress; and as a result, only the seeds which had green lemmas, which are larger and more vigorous, survived the stress.

Inheritance of the Striping Gene (ws ws)

To investigate the mode of the inheritance for this gene, reciprocal crosses of striped mutant X translocation tester stocks and striped mutant X normal were made and grown. When the striped mutant was the pollen parent and the normal genotype used as the female, the F_1 populations were all

Table IX. Seeds from striped mutant produced under different temperatures and their subsequent development into striped and albino seedlings.

Temperatures under which seeds were formed degrees centigrade	No. of Seeds		No. of plants with Seedling Color		Percent Albino
	Planted	Emerged	Striped	Albino	
2-10	35	28	26	2	2
10-15	82	70	43	27	38.5
21-27	102	81	40	41	50.6

Interaction X^2 for albino seedlings = 16.5, $P<.01$.

green; but in its reciprocal cross when the striped mutant was the maternal parent and the normal Titan was the paternal parent, the F_1 population consisted of 54 green seedlings to 31 albino seedlings, a definite indication of extranuclear inheritance for albinism (Table X).

The F_2 population of both types of crosses segregated in a three green to one striped ratio, no albinos were observed in any of these F_2 populations (Table X). One reason why there are fewer striped plants in the F_2 populations than the expected could be attributed to the difficulty in the classification of some plants for the trait.

F_2 heads from normal green X striped mutant were planted in F_3 hills; the F_3 lines were green, segregating (3 green to 1 striped) and mixed striped and albino seedlings on a 1:2:1 ratio, respectively.

The pattern of segregation for the mutant gene suggests that, first of all, the gene is recessive to its dominant allele and upon homozygosity causes the partial mutation of the plastids which results in striped leaves. These mutated plastids self-replicate and give rise to plants devoid of normal green plastids, i. e., albinos. These albino seedlings only survive for a few days until their food supply from the seed is depleted. Striped plants which contain some normal plastids continue their normal life. Sô, as cited by Imai (5) for barley, Imai (5) for barley, Arnason (3) for barley, Pal (15) for rice, and Rhoades (18) for corn reported similiar gene action in their experiments with plastid mutations.

Plastids, being one of the few self-replication organelles inside the cell, have the genetic material of their own to code for their structure during the duplication process. The DNA material inside the plastids are

Table X. Inheritance of striped mutant in reciprocal crosses with normal Titan and Betzes.

Cross	Generation	Number of Plants			Ratio Tested	Chi Square	P
		Green	Striped	Albino			
Striped mutant X normal	F_1	54	0	31			
Striped mutant X normal	F_2	674	201		3:1	2.12	.20-.10
Normal msl msl X striped mutant	F_1	70					
Normal msl msl X striped mutant	F_2	1079	329		3:1	1.95	.20-.10
Betzes ms8 ms8 X striped mutant	F_1	59					
Betzes ms8 ms8 X striped mutant	F_2	148	31		3:1	5.62	.02-.01
Normal msl msl X striped mutant	F_3	189	344	216	1:2:1	6.82	.05-.02
Betzes ms8 ms8 X striped mutant	F_3	20	33	6	1:2:1	7.33	.02-.01

F_2, Sum of X^2's = 9.69, X^2 for all samples: 6.4 heterogeniety X^2 = 3.29, P=.10-.05

F_3, Sum of X^2's = 14.15, X^2 for all samples: 4.04 heterogeniety X^2 = 10.11, P<.01.

called plasmogenes or plastogenes. The mutant gene (ws ws) when in a homozygous condition, causes the mutation in plasmogenes of the plastids, which cause it to lose its normal ability to manufacture chlorophyll and appear white. During the multiplication process, the normal and abnormal plastids replicate and give rise to variegated leaves. When these variegated plants are selfed, the cells which contain only the mutant plastids may give rise to seed and subsequently albino seedlings.

The fact that (ws ws) is a mutator gene may also be indicated in our F_3 hills, where a new mutation of plastids had occurred which resulted in a creamy band on the leaf which persists till maturity.

Location of the Mutant Gene ws ws

X^2 goodness of fit test for the monofactorial ratio for the Rr, Ii, Ee, Vv, Ms8 ms8, Ws ws and Ms1 ms1 genes was applied to F_3 ratios listed in Table XI. Of all the genes listed, only Ms1 ms1, Ms8 ms8, and Ws ws, all in Betzes, fail to fit the proposed monofactorial ratios.

To estimate the linkage intensities between Ws ws, Vv, Ee, Rr, Ii, Ms8 ms8 and Ms1 ms1 genes, a X^2 goodness of fit test for random-assortment of these genes was applied. A P level above 5% for a 9:3:3:1 ratio is considered to indicate independent assortment for two genes in the F_2 population, Table XII.

Since genes Ws ws vs. Ms1 ms1 indicated a possible linkage on F_2 data, further investigation concerning their linkage was determined utilizing complete F_3 classification. Linkage intensities concerning genes Ms1 ms1 and Ms8 ms8 vs. Ee, Ii, Rr, and Vv were investigated in F_3 population and

Table XI. Segregation for Rr, Ii, Ee, Vv, Ms8 ms8 and Ms1 ms1 genes on F_3 hills.

Gene	Generation	Ratio Tested	Observed Ratios AA	Aa	aa	X^2 goodness of fit	P level
Rr	F_3	1:2:1	31	67	36	.51	.80-.70
Vv	F_3	1:2:1	38	74	23	4.54	.20-.10
Ee	F_3	1:2:1	36	63	36	.73	.70-.60
Ii	F_3	1:2:1	33	77	25	3.53	.20-.10
Ms8 ms8 in Betzes	F_3	1:2	48	28		30.1	<.01
Ms1 ms1 in Titan	F_3	1:2	261	489		.72	.50-.30
Ms1 ms1 in Betzes	F_3	1:2	30	29		8.11	<.01
Ws ws in Betzes	F_3	1:2:1	52	72	11	25.4	<.01
Ws ws in Titan	F_2	3:1	1079		329	2.01	.20-.10

Table XII. Calculations of linkage intensities of Ws ws, Vv, Ii, Ee, Rr, Msl msl and Ms8 ms8 from F_2 data.

Aa vs. Bb	Phase	Observed Ratios Genotype A-B-	A-bb	aaB-	aabb	Sum	X^2 for goodness of fit to 9:3:3:1 ratio	P Level	Linkage X^2	P Level	Conclusion
Ws ws vs. Vv	C	104	19	8	3	135	27.38	.01	2.07	.20-.10	Independent
Ws ws vs. Rr	C	88	36	8	3	135	11.03	.05-.01	.23	.70-.50	Independent
Ws ws vs. Ii	R	105	19	8	3	135	28.08	.01	2.14	.20-.10	Independent
Ws ws vs. Ee	R	91	33	8	3	135	20.5	.01	.02	.90-.80	Independent
Ws ws vs. Msl msl	R	766	313	255	74	1408	10.07	.05-.01	5.83	.02-.01	Linked*
Ws ws vs. Ms8 ms8	R	64	26	11	6	107	6.21	.10-.05	.05	.90-.80	Independent
Vv vs. Ii	R	93	18	18	6	135	8.85	.05-.02	1.25	.30-.20	Independent
Vv vs. Ee	R	85	28	15	7	135	5.82	.20-.10	.29	.70-.50	Independent
Vv vs. Rr	C	86	27	12	10	135	8.74	.05-.02	2.87	.10-.05	Indep.**
Ee vs. Ii	C	81	17	30	7	135	4.18	.30-.20			Independent
Ee vs. Rr	R	71	27	26	11	135	1.18	.90-.80			Independent
Ii vs. Rr	R	80	33	18	4	135	6.93	.10-.05	1.12	.30-.20	Independent

*, ** = Recombination value of 45.2% ±1.4, recombination value of 50%, respectively.
C = Coupling, R = Repulsion

are also listed in Table VIII.

Since the sterile plants of F_2 populations with a genotype of ms8 ms8 and ms1 ms1 were discarded, then to estimate linkage intensities of these genes with Ws ws, Ee, Rr, Ii, and Vv genes, which had only been read in F_3, we calculated a X^2 goodness of fit test for a 1:2:1:2:4:2 ratio which indicates a segregation of 2:1 within 1:2:1. A P level above 5% for the calculated X^2 is considered to indicate random assortment and a low P level below 5% indicates a lack of fit to the ratio tested and a possibility of linkage of the two genes exists. Since the number of observed Ms8 ms8 and Ms1 ms1 plants on F_3 hills involving crosses with Betzes did not fit the proposed 2:1 (monofactorial) ratio, then this gene fails to fit the proposed ratios for independent assortment when it is studied with the above mentioned genes. Calculation of the interaction chi-square or linkage chi-square permits a test for linkage freed of the abnormal mono-hybrid ratios. Applying this test showed that in only 5 of the 9 families was there a possibility of linkage. Table XIII indicates a possible linkage between Ms1 ms1 and Ws ws, Ee and Vv and between Ms8 ms8 and Ii and Rr. Calculated recombination values for Ms1 ms1 vs. Ws ws, Ee, and Ms8 ms8 vs. Vv, and Ii are so large that they approach nearly a 50% recombination value which is an indication of independent assortment for these genes. Calculations of linkage intensity of Ms1 ms1 vs. Ws ws from combined F_2 and F_3 data yielded a recombination value of 47.64 with ±1.22, thus indicating a possible independence for these two genes. Application of Allard's homogenuity test indicates that data for these two generations are homogenous.

Table XIII. Goodness of fit chi-squares to 1:2:1:2:4:2 ratio, linkage chi-squares concerning genes Msl msl vs. Ws ws, Vv, Rr, Ii, Ee, and Ms8 ms8 vs. Ee, Ii, Rr, and Vv, F_3 data.

Genes Aa vs. Bb	Phase	Genotype Frequency AABB	AABb	AAbb	AaBB	AaBb	Aabb	Sum	Goodness of fit X^2	P Level	Linkage X^2	P Level	Recomb. Percent and S.E
Msl msl vs. Ws ws	R	79	112	81	114	224	134	744	15.5	<.01	7.96	<.01	49.6 ±1.2
Msl msl vs. Ee	C	8	15	7	9	9	11	59	11.72	.05-.02	2.88	.10-.02	R>50
Msl msl vs. Ii	R	10	16	4	9	13	7	59	12.33	.05-.02	.69	.50-.30	
Msl msl vs. Rr	R	8	16	7	8	13	7	59	10.13	.10-.05	1.51	.30-.20	
Msl msl vs. Vv	R	9	17	5	5	22	7	59	16.19	<.01	3.54	.10-.05	R>50
Ms8 ms8 vs. Ee	C	14	224	10	6	14	8	76	31.6	<.01	.79	.50-.30	
Ms8 ms8 vs. Ii	R	12	32	4	3	16	9	76	46.3	<.01	12.7	<.01	R>50
Ms8 ms8 vs. Rr	R	8	21	19	5	18	5	76	42.2	<.01	11.59	<.01	37.4 ±2.02
Ms8 ms8 vs. Vv	R	15	26	7	7	13	8	76	35.9	<.01	1.27	.30-.20	

** Combined data of F_2 and F_3 populations indicate recombination value of 47.64 with S.E. ±1.22 for genes Msl msl vs. Ws ws.

C = Coupling, R = Repulsion

Translocation stocks were used to study the location of the gene Msl msl. Segregation of families into homozygous fertile and segregating male sterile with a 1:2 ratio, indicates independent assortment of the gene from translocation breakpoints while complete linkage of Msl msl with translocation breakpoints should only yield fertile heads in fertile hills and male sterile heads in semisterile hills. It is concluded that the gene Msl msl is linked with T3-5b and since T3-7a indicates independence of the gene from the translocation breakpoints, then one could propose that the gene Msl msl is linked with chromosome number five. See Table XV. This is in agreement with the findings of other workers (14).

Translocation stocks were used to investigate the location of the gene for striping. A X^2 goodness of fit test was applied to test the linkage of the striped mutant gene (ws ws) with the translocation breakpoints. The results are recorded in Table XV. If segregation of hills into striped, segregating and green fit the proposed 1:2:1 ratio within both the fertile and semi-sterile class, then the ws ws gene is independent of the translocation points. Complete linkage of ws ws gene with translocation breakpoints, the fertile hills should only yield striped and green hills; and the semisterile hills should only yield segregating hills. The data in Table XV indicates a possible linkage of gene ws ws with T3-5b.

As we have indicated on Table XIV, the Msl msl gene is located on chromosome number 5, and our linkage studies suggest possible independent assortment between ws ws and Msl msl genes. Considering the above stated

Table XIV. Linkage of Msl msl with translocation breakpoints. F_3 Hills.

Translocation Stocks	Ratio Tested	From Fertile F_2 Heads: No. of Families Fertile	Segregating	X^2	P Level	From Semi-Sterile F_2 Heads: No. of Families Fertile	Segregating	X^2	P Level
T1-7a	1:2	33	67	.001	.99-.95	22	78	5.72	.02-.01
T2-4a	1:2	15	18	2.17	.20-.10	2	15	3.54	.10-.05
T2-6a	1:2	40	44	7.71	.01	21	35	.42	.70-.50
T3-5b	1:2	45	17	42	.001*	11	77	17.1	.01**
T3-7a	1:2	39	78	0	1	35	47	3.25	.10-.05

*, ** = Recombination value 10.5 ±2.7 and 12.8 ±3.8, respectively.

Table XV. Segregation pattern of hills in F_3 population from:

Trans-location	Class	Ratio Tested	From Fertile Heads Observed	X^2	P Level	Recomb. Percent and S.E	From Semisterile Heads Observed	X^2	P Level	Recomb. Percent and S.E
	Striped	1	24				29			
T1-7a	Segregating	2	48				48			
	Green	1	28	.49	.80-.70		23	.88	.70-.60	
	Striped	1	10				6			
T2-4a	Segregating	2	16				9			
	Green	1	7	.48	.80-.70		2	1.94	.50-.30	
	Striped	1	25				18			
T2-6a	Segregating	2	42				29			
	Green	1	17	1.52	.50-.40		9	2.9	.30-.20	
	Striped	1	20				19			
T3-5b	Segregating	2	16				44			
	Green	1	24	13.59	P<.01	14.8 ±3.1	27	1.45	.50-.40	R>50
	Striped	1	34				23			
T3-72	Segregating	2	49				34			
	Green	1	34	3.08	.30-.20		26	3.9	.20-.10	R>50

findings, one could propose that gene ws ws is either on chromosome number three or five. Additional populations need to be evaluated.

Allard's handbook was used (1) to facilitate the calculations of recombination values.

SUMMARY

A striped mutant of Titan barley which arose spontaneously and upon selfing yielded striped and albino seedlings was the subject of this study.

Marked seeds with predominately albino lemmas gave rise to a higher frequency of albino seedlings than seeds formed within green lemmas and paleas. It was observed that small seeds yielded predominately albino seedlings while large ones gave rise to predominately striped seedlings.

Seeds of the striped mutant raised at various locations were planted and their segregation into albino and striped seedlings were investigated. There was a definite difference in segregation of seeds from various sources and this difference is attributed to the environment under which the seed was grown.

To investigate the effect of temperature on the amount of striping, seedlings of the striped mutant were raised under various temperatures and it was observed that seedlings raised under colder temperatures display more striping than the ones grown at warmer temperatures.

Striping of the first leaf was a function of temperature at the time the seed to produce the first seedling leaf matured, whereas the subsequent leaves were more influenced by the current environment.

In comparison to normal Titan, it was observed that the seed from the mutant type had a higher average agtron reading (was lighter in color) and had a higher percent protein than normal Titan.

The differences in kernal weight and percent plumpness of the mutant type and normal Titan were nonsignificant.

Yield trials of the striped mutant in comparison to normal Titan were conducted at several different locations. The mutant type was lower in yield than the normal Titan in all trials.

To investigate the mode of inheritance for the striping gene, reciprocal crosses of the striped mutant X translocation tester stocks and striped mutant X normal Titan were made and grown. It was concluded that the albinism is transmitted cytoplasmically. Further investigation has revealed that the gene for striping is recessive to its dominant allele and upon homozygosity, it causes partial mutation of the plastids which results in striped leaves. These mutated plastids self-replicate and give rise to plants devoid of normal green plastids, i. e., albinos. Translocation stocks were used to study the location of this recessive gene, and it was concluded that this gene is probably located on chromosome five with a possibility it may be on chromosome three.

The gene was determined to be independent of the breakpoints on translocation stocks T1-7a, T2-4a, T2-6a, and T3-7a, but linked with T3-5b. The gene was determined to be inherited independently of genes on chromosomes 1, 2, and 7 but to be possibly linked with msl on chromosome five.

Crosses of missing translocation stocks (T1-6c and T4-5a) X striped mutant and the reciprocal crosses of striped mutant X translocation stocks were made to confirm and further study the location of the gene for striping.

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