

The discovery of Ds. Its behavior and its relation to mutable loci.

I. Review of previous talk:

1. Sudden appearance of a large number of newly arisen mutable genes -- expressing variegation-- in progeny derived from self-pollinations.

2. The origin in plants that had undergone the chromosome type of breakage-fusion-bridge cycle in early development. Healed broken ends of chromosome 9 in recovered branch.

3. The various types of plant characters showing variegation. Various types mentioned affecting chlorophyll expression. More types later affecting other characters.

4. About 40 different origins of mutability recognized in early experiment

5. The features shown by the seedling exhibiting variegation:

a). Background - altered phenotype.

b). On background, streaks of normal phenotype

c). The normal streaks present in a decided pattern. Reflect:

(1) Time of change to normal during development of tissue -- determines the size of the streak

(2) The number of streaks: reflects frequency of occurrence of changes to normal phenotype during development.

(3) The relative distribution of streaks: reflect the distribution among a growing tissue in which changes occur in specific cells.

(4): The different types of patterns: The Seedlings:

Many later occurring changes



Few late occurring changes.



any early changes. Leaf a mosaic of many patterns.



d). Within a single progeny, one type of pattern usually predominant.

6. The changes in the pattern of mutations occurring during development.

a). Sectors formed: 1). ↑ increased number of streaks
2). Decreased number of streaks
3). No streaks
4). Large mutant sector

b). The twin-sectors: One sector composed of two parts:

- Types: 1). Normal phenotype: altered phenotype - no streaks
2). " " : " " - increase or decrease
in number of streaks.
3). No streaks : increased or decreased frequency of streaks.
4). Increased frequency of streaks: decreased frequency of
streaks.

c). The interpretation of the twin sectors: Each component of twin derived from one of two sister cells. Both sectors arose from single cell as consequence of a mitosis, ~~event~~ which produced two sister cells.

7. Interpretation of the nature of the change that occurred to give rise to single or twin sectors:

a). Particular pattern in seedling leaf reflects a controlled type of expression of time when mutations will occur, and the cells in which they will occur. *at particular stage or during tissue*

b). The factors responsible for this control may be altered as a consequence of a mitotic event.

c). Following alteration, there will be a different pattern of mutation in the descendent cells from that present before the change occurred.

d). The twin sectors suggest that controlling factors segregate at a mitosis: one cell receives something that the sister cell lost.

3). The mutant - altered pattern twin sectors: Suggest that the mutation itself is consequence of a mitotic event altering something.

8. The major questions and propositions:

1. What factor or factors are responsible for producing a particular pattern of mutations during development? i.e., (1) the time during the development of a tissue when mutations will occur. (2) The cells of the tissue in which this will occur; i.e., the frequency and distribution of changes that will occur in particular cells during development.

2. How does the factor, or do the factors segregate in certain cells as the consequence of a mitotic event?

3. Is the process of mutation associated with the same type of event?

4. What component of the mitotic process is involved?

a). Is it a segregation of cytoplasmic components?

b). Is it related to the chromosomes? To changes at specific loci?

c). Is it related to the process of chromosome reduplication?

II. The period of decision and the early investigations.

1. Original purpose, to analyse the genic components within the short arm of chromosome 9.
2. The presentation of a wholly unexpected phenomenon - the variegations.
3. Gradual conviction, following growing of the variegated plants -- that the phenomenon involved in the expression of variegation was far more important than that of the original purposes, stated above.
4. How could this phenomenon be investigated?

a). At first, did not know.

b). As first step, decided to investigate inheritance behavior of a number of selected cases; and the inheritance aspects of the pattern controlling systems. This in order to get facts with which to think.

5. First year and a half -- Results confirmed earlier observations. Showed that the alterations of phenotype were changes at the locus of a particular gene; a mutable gene. The change resulted in mutation from recessive to dominant. (Captions).

The control of the mutation process not well distinguished in early studies but suspected a similar type of change as that producing mutation.

III. The discovery of a variegation system that altered the chromosome composition of nuclei during development. Its major mode of operation:

1. ~~Witherxxxx~~ A part of short arm of chromosome 9 eliminated in certain cells during the development of a tissue.

2. The pattern of such losses in the various tissues simulated the patterns produced by the mutable genes, where mutation from recessive to dominant occurs.

Losses



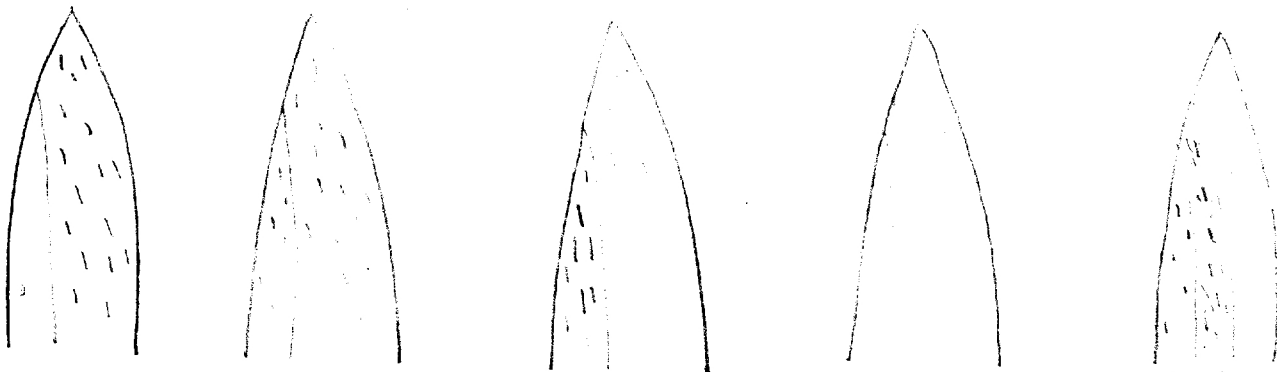
loss of band in white gene, normal

Mutations



*dominant - normal green
recessive - white - variegation*

3. The changes in patterns of loss, as seen in sectors, similar to the changes in mutation time and frequency noted with mutable loci:

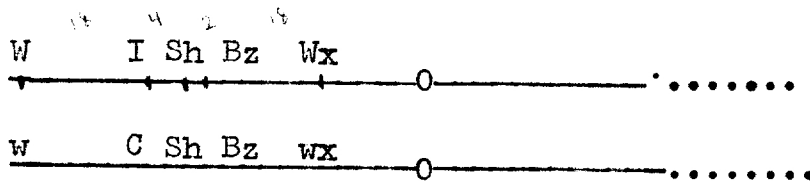


normal frequency

low frequency

IV. How the system involving chromosome loss was first recognized:

1. First seen in some kernels derived from self-pollination of one plant that had undergone chromosome type of b.f.b. cycle in early development
2. This plant -- carried two chromosomes 9 with newly healed broken ends.
3. The genetic factors carried by each chromosome:

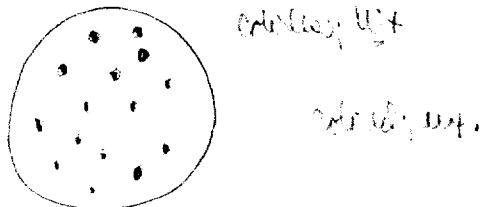


4. The factors carried in the short arm of chromosome 9 -- necessary to know them to continue study:

W, w; Yg, yg. -- At end of short arm.
 I, C, c
 Sh, sh (a) Crossing over between markers
 Bz, bz (b) Physical position in chromosome
 Wx, wx

ILLUSTRATIONS Ears

5. The pattern of variegation exhibited on some kernels of self-pollinated ear:

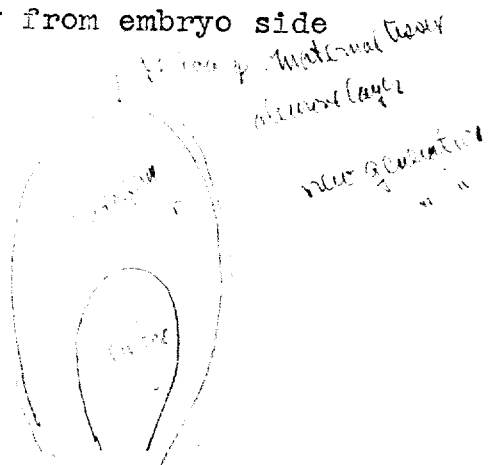


V. The structure of the maize kernel and its development.

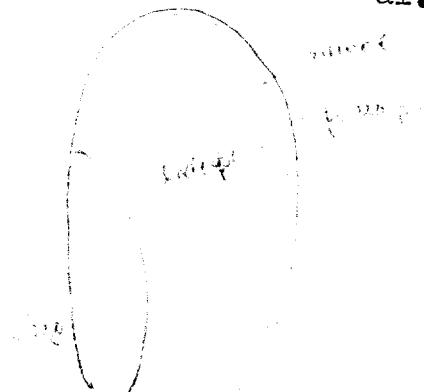
1. Must understand this for any discussion of maize genetics or cytogenetics. Will be given diagrammatically for those who are not familiar with the morphology and development of kernel.

2. The morphology of the mature kernel: (ILLUSTRATIONS * Cut kernels).

View from embryo side



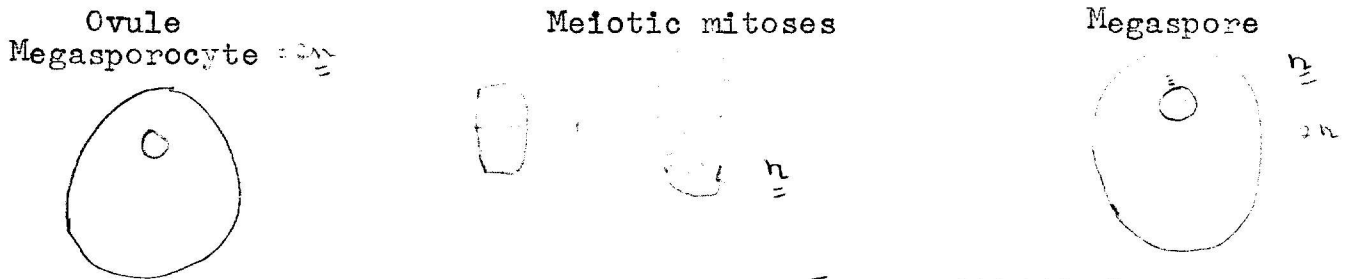
Cross section -- longitudinal.



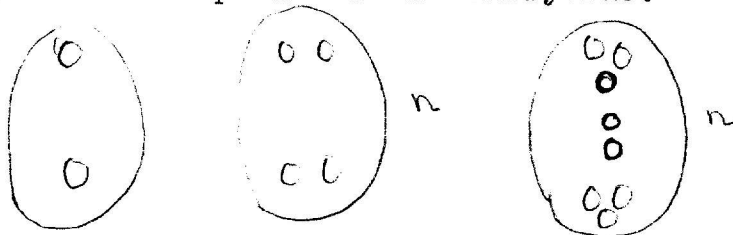
- illustrations
- 1) Randolph
 - 2) Kernel body
 - 3) Cut kernels.

3. The origin of the endosperm and embryo in the kernel.

a). The ovule on ear, before fertilization:



b). The development of the embryo sac: From megaspore n



4. The development of the male gametes:

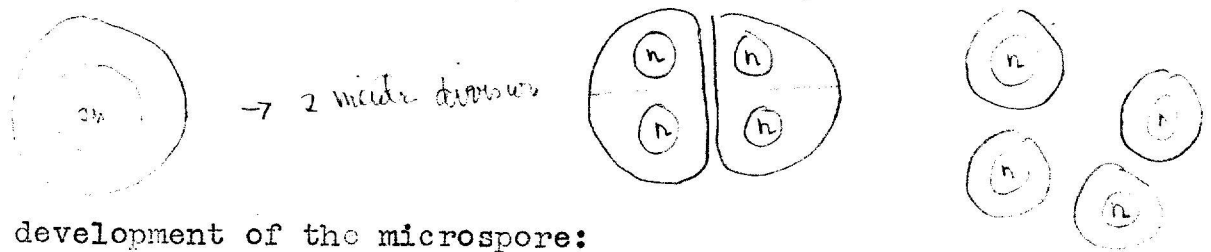
a). The young tassel, inside of leaves: telescoped effect.

b). Composed of flowers along branches of tassel.

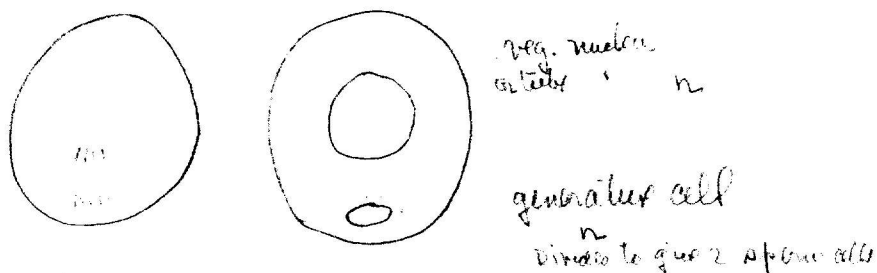
c). Each flower has three anthers.

d). Inside anthers at young stage are enlarging cells - the microsporocytes.

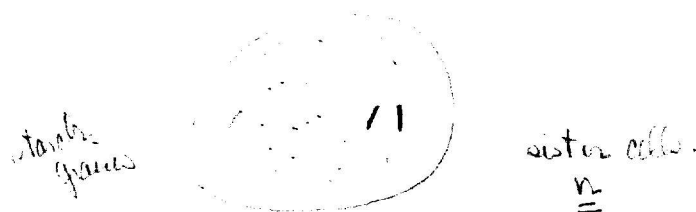
e). The meiotic mitoses and development of microspores:



f). The development of the microspore:

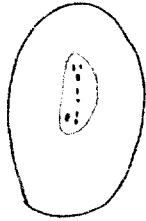


g). The development of the pollen grain and sperm cells:



5. The process of fertilization:

Pollination; pollen tube; break into embryo sac



The nuclear fusions.

The embryo and endosperm develop.



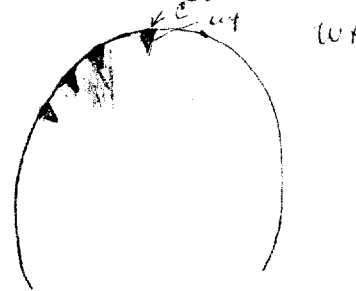
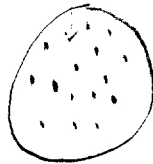
²ⁿ
Zygote, Develops
embryo
Primary endosperm
nucleus: 3n

VI. Return to case of chromosome loss variegation.

The particular kernels

1. Assume female C wx / C wx x male I Wx
2. Loss of I and Wx during development: Areas of color in aleurone.
Underlying areas, exactly corresponding, wx.

3. Cross section of kernel:



4. The pattern of such losses:

- a). Exceedingly uniform in some kernels with regard to size and distribution of recessive spots.
- b). Not like pattern produced by other known mechanisms that result in losses of chromosomes: the breakage-fusion-bridge cycles
Ring chromosome behavior.
- c). Conclusion: A previously undiscovered mechanism at work here that produces the losses of I and Wx simultaneously.

5. Plants grown from the variegated kernels. Showed:

a). White streaks on normal green background:
in some cases,



b). Sectors appeared: no losses, increased or decreased frequency of loss, earlier or later timing of losses.

c). The twin-sectors: No losses -- increased or decreased freq. of loss

Altered pattern of losses in each sector: Often increased in one and decreased in other.

6. Conclusion: The mechanism responsible for loss of a chromosome must be basically similar or the same as that producing the gene mutations.

a). The "loss" corresponded to the gene mutation, recessive to dominant.

b). The system controlling the time of loss, and the cells in which it will occur is the same type as that controlling when gene mutations will occur in the cases of the mutable genes.

c). Somatic segregations, occurring at a mitosis, of the factors associated with the controlling system occur during development and this results in altering the time and frequency of losses in descendent cells.

d). Some reciprocal relationship exists in the two sister cells resulting from this particular type of mitosis -- as if one cell gained something that the other cell lost. in comparison with system present in the mother cell.

7. The summary: Comparisons between chr. loss and mutable loci:

a). Mutable loci: Mutation from recessive to dominant; Not chromosome loss.

System present controlling when these mutations will occur.

Controlling system altered as consequence of a mitotic event. Resulting two cells differ from mother cell.

b). The chromosome loss phenomenon: Very same kind of mechanism involved.

Change occurs resulting in chromosome loss

System controlling when this will occur

This system altered as consequence of mitotic event. Resulting cells differ from mother cell with regard to this.

c). Something happens to chromosome to result in loss. What is it?

d). The loss must be gross to include nearly all of short arm of chromosome 9 as seen in the sectors: Losses of W, I and Wx.

e). Therefore, this mechanism is subject to microscopic examination.

(1) Should be able to find evidence of this loss.

(2) This has been accomplished, and will be described in detail shortly.

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9. Tests conducted to determine whether or not a whole chromosome or only part of chromosome 9 being lost.

a). Example given only. Many different types of tests conducted:

Variegated plant with: $\frac{I^A Sh Bz Wx}{C sh bz wx}$ 1 Ac
crossed as male to tester plant that was C sh bz wx.

Diagram normal crossing over between markers: See below

Resulting plant: $I^A sh Bz Wx$
 $C sh bz wx$

The types of kernels obtained:

- non- variegated: $I sh Bz Wx = 1$ colorless sh Bz Wx: 1 variegated for colorless areas *See photographs.*
- $C sh bz wx =$ all $C sh bz wx$
- Reg. 1: $I sh Bz wx =$ all colorless sh bz wx. non-var.
- $C sh Bz Wx = 1$ C sh Bz Wx non-var. : 1 C sh Bz Wx with *recombination* sh bz wx
- Reg. 2: $I sh bz wx =$ all colorless sh bz wx non-var
- $C sh Bz Wx = 1$ C sh Bz Wx non-var: 1 C sh Bz Wx with *recombination* sh bz wx
- Reg. 3: $I sh Bz wx =$ all colorless sh Bz wx. non-var
- $C sh bz Wx = 1$ C sh bz Wx; no var for *colorless* wx: 1, with *recombination* sh bz wx, non-var.

b). Conclusion: Some factor located to right of Wx, associated with loss phenomenon. This is because Wx locus required for losses to appear. All crossovers up to this locus, bringing in wx, do not show the loss phenomena.

c). Cross of same plant as male to plant that is c sh Bz wx:

$c sh Bz wx \times \frac{I^C Sh Bz Wx}{C sh bz wx}$

Types of kernels expected from non-crossing over and crossing over:

- $I sh Wx \times \rightarrow 1$ colorless sh Wx : 1 colorless sh Wx with *wx spots*
- $C sh wx \rightarrow$ nearly all C sh wx non-var. About 1% of C class but in C sh wx kernels showed *recombination* pattern *See photos*
- $I sh Wx = 1$ C sh Wx : 1 C sh Wx with *recombination* sh wx areas
- $C sh Wx = 1$ C sh Wx : 1 C sh Wx with *recombination* sh wx areas.

