Lecture 1. Cornell University. Tuesday, November 16, 1965

Aspects of Gene Control in Higher Organisms.

I. Influence of bacterial and phage genetic and molecular studies on genetic and biological concepts.

1. Basic genetic manterial: DNA: its bases, its organization; its coding.

2. Mode of operation of the genetic system: DNA Replication- DNA polymerase

DNA transcription; RNA polymerase, base sequence of mRNA, mRNA transcriptions through ribosomes; transfer RNA, associated enzymes. Proteins formed; specificities.

4. Types of genetic components in the bacterial DNA:

Structural genes
Genes for transfer RNA
Genes for structural proteins
Genes for ribosomes: B. subtilis:

5. Regulation of action of genes: Induces and Repressors.

The Operator-- reading frame of structural gene
Regulator genes.

Organization of structural genes: Operons

One Regulator: Positions. Number of genes: their relations

Operon plus others: one regulator. Arginine.

Regulators: adjacent to operator: Lac locus.

The activator component: Regulator: positive: Arabinose

A B D C + E The C: Activator.

The feedback mechanisms.

6. Regulation at the gene level: not yet clear how this operates.

7. The forgotten type of regulation: The H_1 and H_2 duplicate genes in Salmonella -- will return to this later.

The host range: Modification of phage genome by host.

8. Effects of episomes: Isolated episomes; the controlling episomes (Austin) (Dawson).

9. IMPORTANT: The acceptance of differential regulation of gene action: enormous stimulus to reconsider mechanisms responsible for control of gene action during differentiation of higher organisms.

Although the basic components are the same: DNA etc, the mechanisms controlling the action of the genes in higher organisms are far more complicated. This related to the very highly organized, complex bodies, the chromosomes, and the highely organized nuclei.

The extraordinary complex organization of the chromosomes and nuclei probably related to mechanisms controlling the action of genes during development.

The various components of the chromosomes and the organized nuclei probably represent the components behaving like those of a computor: highly organized set of ~~con~~sequ~~e~~tive events, each related to the previous event:

Example of this: ⓐ One must consider the caterpillar and the moth: Both are extraordinary complex individuals but utilize the same set of genes.

ⓑ One must consider polymorphism: mimicry patterns, etc.

The computer: the "Swith Genes": One or Two mendelizing units switch the computer from one sequence of interplay of events to another sequence, using the same complement of genes for this.

10. We must consider that regulation of gene action in a higher organism is a highly programmed sequence of events from egg to mature individual. The large number of different components of the chromosome are probably the component elements in this programed sequence. Question: What do we know about these components of the chromosome and what do we know about individual mechanisms in the system?

II. The Chromosomes and their component parts:

1. Compared to bacterial chromosome, the breadth of the chromosome is enormous: clearly visible in the light microsoope.

2. Number of DNA molecules within an individual chromosome: Very recent investigations, taking DNA from the chromosomes, suggest that each chromosome has relatively few DNA molecules. (8 to 10?)

3. The non-DNA components of the chromosomes:

a). The Histons: Ten different types known. Fall into 4 general classes:

Ia, Ib, II, III, IV. Based on lysine-arginine ratio
8:1 10:1 1.7:1 0.7:1 0.7:1

(No tryptophane)

Same qualitative types of histones in all nucleated organisms.
" " " " " in active and in inactive chromosomes with very few exceptions.

Some turn-over of histones in nuclei that do not replicate DNA

Quantitative differences in histones but not qualitative differences.

Considerable amount of evidence that histones related to repression of gene action. But, what substances are related to activation of gene?

b). RNA, new species, related to histones. One of these RNA molecules for several of the histones.

c). The residual protein - acid type; Phosopho proteins.

RNA, new species, associated with this protein.

d). Phosopholipids.

4. The chromosome: an extraordinarly complex system: Related to control of action of genes; to mechanisms producing mRNA etc. and to mechanisms of reduplication of the chromosomes, etc.

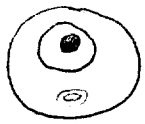
We should expect to find that new, previously unsuspected mechanisms of protein formation, and replications may occur with these structures.

5. Bacterial systems: do not have all of these. A few exceptions indicated and these may help in explaining some of the modes of regulation of gene action in higher organisms. The one outstanding example is the control mechanisms operating at the H_1 and H_2 duplicate genes in Salmonella.

III. The relation of the organization of the chromosome parts in the nucleus(to gene action and repression.) GENERAL CONSIDERATIONS.

1. The active chromatin: dispersed; the inactive chromatin contracted.

a). This realized by cytologists for many years: Example in the Microspore of plants:



b). Condensed nuclei and chromatin in generative cell; size of nucleolus.

2. The nucleolus organizer: Special region in one chromosome. Function: to produce the nucleolus at telophase; The nucleolus with the DNA of organizer: related to production of the cytoplasmic ribosomes.

Example of the organizer: its position with respect to the nucleus; the constance of this position in the working nucleus.

Slide 1: Maize set of chromosomes at pachytene.

Slide 2: The nucleolus chromosome in maize.

3. The true heterochromatic regions within a chromosome.

a). The heterochromatin about centromeres in Drosophila. The chromocenter in the working nucleus. Its position with reference to the nuclear membrane.

b). The true heterochromatin in maize: the knobs: Slides 3.

c). The relation of the knobs to the nuclear membrane: Slide 4

d). The relation of the centromeres to the nuclear membrane: Slide 5

e). The heterochromatin at the ends of chromosomes: Very visible in many plants and animlas: Relation to nuclear membrane.

4. None of these types of "heterochromatin" have conventional genes in them.

Imp. 5. The relation of chromatin parts to the nuclear membrane is highly significant as will be indicated.

carrying conventional Feulgen

IV. Condensation of chromatin: one form of control of action of genes.

Two distinctly different classes of this; each , however, is effective in repressing the action of structural genes.

CLASS I:

1. Two nucleated microspore: The active nucleus and the inactive nucleus. organization of chromatin in each.

2. Types of condensation of chromatin in different nuclei of the same tissue: rabbit retinal cells in embryo; Beermann photo. Slides 6, 7.

3. Calf thymus nucleus: Frenster, Allfrey, Mirsky Lab.

a). Appearance of nucleus in light microscope before and after swelling. Slide 8. Position of condensed chromatin in condensed regions Slide 9.

b). Tests of parts of chromatin in such nuclei that produce mRNA: Slide10

c). The relation of strands of DNA in association with condensed regions: Figure of Frenster: Draw.

d). Components of chromatin in active and inactive chromatin: Table 1, Frenster May, 1965:

	Ratio Active Chromatin / Inactive
Histones	0.90
non-histone residual protein	2.00
Phospholipids	4.93
RNA	5.15
Phospho-protein phosphorous	3.74

e). Effect of additions of polyanions to active and inactive chromatin:

Suggests that the special species of RNA associated with the residual DNA protein associated with activation of gene whereas RNA species associated with histones may regulate in some manner repression of DNA.

4. The contracted chromatin: Specific types for each type of cell.

5. QUESTIONS: (1) What type of mechanisms controls these highly specific types of contractions of chromatin?

(2) Why are the condensed parts bulked as they are with the active parts extending from them in all parts of the contracted mass?

(3) What is the relation of the nuclear membrane to all of this? How does it come into the picture of repression?

(4) What types of determinants control the particular parts of a chromosome that will be in the condensed stage at any one time or in any one type of cell?

(5) What components of the chromosomes are "set" in advance so that they will become contracted in certain cells? What

Ish.

mechanism serves to release these settings? Are they particular elements associated with the DNA-- the genes? and if so, what are these components?

(6) If (5) is in fact correct, do we have any evidence for such settings and erasings of a setting?

These questions indicate the extent of our ignorance of the control mechanisms in nucleated organisms at the molecular level. We are not yet ready to consider in detail any control mechanism in higher organisms at this level. We must know more about the specifics of chromosome composition, organization of parts, and changes in this that occur.

CLASS II CONDENSATIONS. This involves the completed contraction of an entire chromosome or of a continuous segment of an entire chromosome in contrast to Class I which involves intermittent condensations within a chromosome.

1. Our knowledge of the control mechanisms better with this class.

2. The X chromosome in mammals as an example of this:

(a). XY : X chromosome is not contracted.

(b). X X: One X contracted; other X not contracted.

(c). Position of the contracted X in the nucleus: At the nuclear membrane. Slide 11

(d). Contraction of X when more than two present:

XX, XXX, XXXX, XXXXX

(e). Selection of which X to contract: XX female: Occurs during development: one X in one cell, the other X in another cell.

(f). How is this selection accomplished? remainsact Why onely one. Will consider this shortly after several other cases reviewed.

(g). The CONTROLLING ELEMENT responsible for contraction allong the chromosome.

(1). Translocations between X and autosomes in the mouse: Russel.

A region of control in the X chromosome.
Contraction occurs to either side of it. [spreading effect]
Distance controlled by this region
Translocations: autosome genes in line with this, they become contracted and non-active:

Diagram:

Contraction: Related to control of one region?

(2). Control of selection process in mammals: will consider after discussion of other cases. Will make discussion clearer.

3. The condensation of a whole set of chromosomes: the mealy bug. Coccid.

(a) Male germ line: One set from father: contracted set
One set from mother: not contracted.

(b) Sperms: Set from father discared during meiosis; only set from mother is carried in the sperm.

(c). The female: regular meiosis:

Some females: produce eggs that develop into females.

Under some conditions, produce <u>some</u> eggs that develop into males.

(d). The female embryos: both sets of chromosomes are functional and euchromatic.

(e) The male embryos: One set--that received from father, carried in the sperm-- becomes totally condensed: Slide 12. Contracted chromosomes again up against the nuclear membrane. Setting to do this occurred in germ line of male: then euchromatic.

(f) Another group of coccids: Same general conditions up to time of condensation of male set in the male embryos. Instead of condensation of set, all chromosomes are eliminated from the nuclei at a certain cleavage division.

(g): Conclude: some relationship between condensation and elimination process as these are related in evolution. Same components controlling both must be involved.

4. The E for elimination chromosomes in the cecidomyidae:

(a). Egg after fertilization:

(b). The pole plasm:

(c). Nucleus associated with pole plasm(germ line). Rescued from elimination. Any nucleus placed here: no elimination will occur. Elimination will occur if nucleus normally here replaced by one that would otherwise form soma and have chromosomes eliminated.

in <u>all</u> nuclei

(d). Chromosomes set for elimination at particular division in cleavage.

Resuce from this setting by contact of nucleus with particular cytoplasmic component.

(Must keep the setting in advance for elimination and rescue of this by cytoplasmic component in mind for later use with X chromosomes in mammals: a rescue process.)

5. To get all of this in focus, will consider the case of Sciara: Controls of the behavior of the X chromosome:

(a). The germ line of the male: 4 chrs. from mother, 4 from father at <u>late</u> stage in spermatogenesis.

(b). Spermatogenesis: Meiotic divisions:

(c). The two types of females: All with normal meiosis

X'X = produces eggs, all of which develop into females.

XX = produce eggs, all of which develop into males.

(d). The zygotes and early cleavage nuclei:

Eggs from X'X individuals: Cleavage:

Zygote: X A mother/ X X A father

Cleavage: 6 or 7th division, one X from father eliminated. Embryo develops into a female.

Eggs from X X mothers:

Zygote: X A/ X X A: Cleavage: both X chromosomes from male eliminated: develops into a male.

(e) Translocations between autosomes and X. Various ones worked with.

Result: The X chromosome:

All the aberrant events controlled by some element carried in the tiny short arm of X which is heterochromatin. If translocation occurs in short arm between centromere and heterochromatin, the chromosome carrying this and all of the autosome part: undergoes same events as X chromosome.

The Heterochromatin carries a controlling element that accomplishes all of the events. How does it do this?

(f). Transloc tions with aberrant disjunctions:in the females:

Female producing females:
X^{g}/X^{t} mothers: Few Eggs: No X; only autosomes.

Constitution of zygote: OA A/ X X A

Elimination: One chromosome X only

Translocation from XX^{t} mothers: Normally male producers

Some eggs: Two chromosomes with controlling element of X

Constitution of zygote:

Elimination: two X chromosomes in cleavage(from father).

Exceptional females: retain two X chromosome as one particle

(g). Suggests: The X (not the X') produces somes substance in egg before meiosis that can rescue one X from elimination With XX females; two such particles. Therefore 2 X chromosomes may be rescued by it.

6. The relation of Sciara evidence to X chromosome condensations in mammals.

   a). All X chromosomes are set in advance to become condensed during development of embryo.

   b). Can be rescued from this by reaction with some component in the cytoplasm.

   c). Only one such component produced.

   d). This element reverses the setting for condensation.

   e). Should be matter of chance which X, if more than one present, will be rescued during stage when rescue occurs.

   f). XY - the X always rescued as no other X present.

   XX - One X rescued in some cells, other condenses; In other cells, the other is rescued.

   XXX, XXXX, XXXX - only one X can be rescued; all others are condensed.

7. Return to Sciara or the coccids: Setting of elements must occur in the germ line of the father.

   Erasing of the setting must occur in the germ line of the mother.

8. Certain chromosomal components, Elements are responsible for the control of of contraction and release from contraction; for repression and release from repression.

   These elements respond to cytoplasmic or intranuclear substances, previously produced or introduced: as with hormones that react with the chromosomes themselves-- special parts.

9. If we accept that there are chromosomal elements, distinct from the genes, that control their action through various types of responses of the chromatin materials, we are a long way on the road to understanding the mechanisms that operate in higher organisms to control the action of genes.

V. Other evidence of the manner by which the action of genes are controlled:

1. Lactic dehydrogenase genes: H and M. Tetrameres:
   HHHH, HHHM, HHMM, HMMM, MMMM, Young and older individuals.
   (Chick; rabbit)

2. Intra-allelic repression: Tetrahymena and Paramoecium

3. Multiple genes - serotypes in Paramoecium: Only one active at a time; others inactive.

4. Esterase alleles in maize: Control of action of one or the other allele: Differences in different tissues.
   Genetic analysis: Control mechanism associated with something at the locus of the gene.

5. Setting of a gene locus at one specific time in one type of cell to be expressed in cells some cell generations later:

   Position effect: Becker w^m locus. Adjacent to heterochromatin by translocation.

   Gene set in some cells at this time. Effect of extra Y: No effect on the time of setting. Effects frequency of cells that have their locus set at this time.

   Reflects type of setting mechansims that occur during development.

6. Alleles that control the pattern of distribution in a tissue of the end product of sequence of gene actions: Pigment as example.

   Lady Beetle: Pigment patterns in the Elytra.

   Types of patterns observed in nature.

   Genetic analysis: One locus associated with control of this pattern. Each allele of this locus responsible for one particular pattern: for control of production of end product of genes in sequence to pigment formation.

   Combination of two alleles of this "regulator" element

   Slide 13 Overlapping patterns.

   Each expresses itself independently of the other. Thus, cytoplasm in this case is not a determining factor directly. Time of gene action of an allele, number of cells in which action will occur controlled by some component at this gene locus.

7. Lady Beetle patterns: Can duplicate completely with maize where we know the element at the gene locus and how it operates to accomplish these patterns. This will be considered in next lecture.

VI. Conclusions: There are particular chromosomal components, chromosomal elements, that are responsible for the control of gene action and they accomplish this in various manners: contractions and release from contractions; repression locally and release from this repression.

The Organization of the components of the chromosome, their numerous components, are all involved in the orderly control of this.

QUESTION: What are the elements that are localized at the different structural gene loci? What are the elements composing the true heterochromatin? What is the nature of the setting and release from setting of such elements? These are some of the basic questions that must be solved in considering mechansisms in organisms with true nuclei that control the action of the genes.

QUESTION: How many kinds of elements are there that do these controls?

My conviction: Relatively few such elements. Like a computor -- not so many different types of elements, but how these are integrated into a programming system. Some basic simplicity, as with structure of DNA.

Illustrations: The Mxxxxxxx Caterpillar and the Moth: same genome but different programing of gene action.

The Switch genes: Polymorphism; Mimicry. Only one or two mendelizing units responsible for altering the programming.

VII. Subject of next lecture: Nature of elements that control the action of specific genes, their independence of the structural genes, the variety of modes of control of gene action thatone element can produce, the extraordinary economy of such elements -- many different genes may be controlled quite differentially by one system of elements. The indication that such elements reflect some basic symplicity in pattern of action just as DNA has a basic simplicity of component parts that can lead to tremedous diversity of effect.

1. The elements in maize initially discovered because they could (but need not) transpose from one location to another in the chromosome complement.

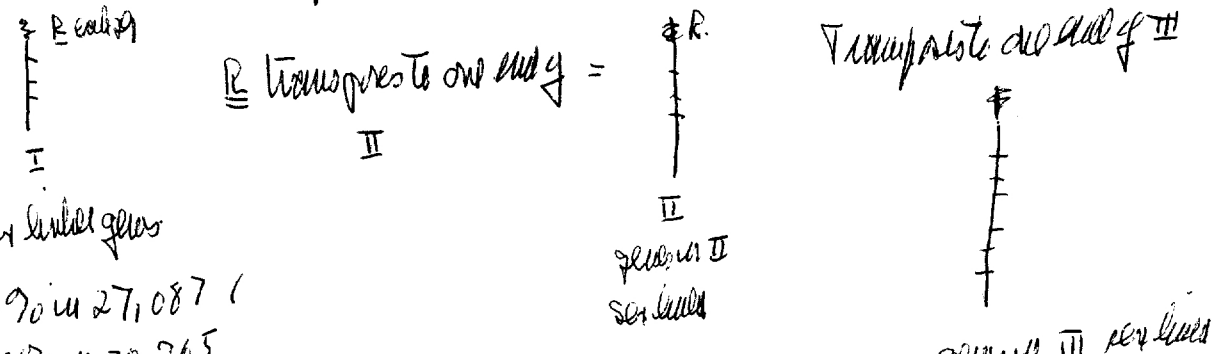
2. Literature: many examples of phenotypes that resemble those produced by the controlling elements in maize but no instances that are supported of the elements that are responsible for this.

3. The transposition of elements to different sites in the genome:

Bacteria: the episomes.

Higher organisms: Only one case of "transposition" of some component and this one that contrlls sex. [Possibly similar in Ephestia]

Sex control mechanism in Megaselia - a fly:

1. 3 pairs of chromosomes I, II, III
2. Genetic markers in each -
3. Sex determiner - component at end of one element -- "sex factor" - on I
4. 

From
III to II or I = 0.38% in 27,087 (
II to I = 0.05% in 30,765
I to II = 0.05% in 23,007
I to III = 0 " in 25,931 flies

Slides. Lecture 1

1. Chromosome set at pachytene: maize
2. Nucleolus chromosome at pachytene:maize
3. Nucleolus chromosome, knobs, pachytene, maize
4. Knobs at nuclear membrane; endosperm of maize, polyploid.
5. Feulgen stain, pachytene maize; B-type chromosome centromere.
6. Kitten, 10 day old retina cells: Beerman
7. Same
8. Lymphocyte nuclei: non-swollen; swollen
9. " " swollen
10. " Active and inactive chromatin: autoradiograph tests.
11. Sex chromosome in embryo tissue of rabbit.
12. Condensed set of chromosomes in spermatogenesis Cerococcus (mealy bug).
13. Elytra patterns: ~~xxxx~~ Lady Beetle. Tan, 1946

Cornell University, Lecture 1. Tuesday, Nov. 16, 1965

ASPECTS OF GENE CONTROL IN HIGHER ORGANISMS.

I. The influence of bacterial and phage genetic and molecular studies on genetic and biological concepts.

1. DNA - mRNA (r. polymerase) - transfer RNA, ribosomes etc. DNA replication

2. Types of genes: structural, sRNA, rRNA, regulators, suppressors, Operator- operon. Regulation.

3. Regulatory mechanisms: not yet clarified.

4. The odd types of control in bacteria:

H_1-H_2 duplicate genes: Phase variation

Episome control of gene action: Taylor; Dawson.

Host modification of phage: Modification of phage genome - restriction of host.

5. Importance of the "odd" types: particularly the H_1-H_2 duplicate genes.

II. Types of gene control that have been examined: Examples.

1. At the molecular level: Haemoblobin in young and older individuals.

Lactic dehydrogenase: Two genes, M,H, The tetrameres. Young vers older embryos.

Esterases in maize: Timing of action of different alleles: constancy for an allele: Control: genetic methods, resides at the locus of the gene. the special advantage of maize.

2. Paramoecium and Tetramymena: Interallelic repression. Timing of event.

Series of genes for serotype: 14; only one active at a time; other "duplicate" genes turned off.

Other genes: behave similarly. Timing of events: controlled. Changes through environmental changes: the serotypes.

3. The position effect in Drosophila: Becker: white locus next to heterochromatin: timing and frequency of this. Effect of Y. Does not alter time of event; alters frequency among cells in which event occurs.

4. The proteins of the 7S component of antibodies: amino acid differences in one segment of this component.

5. Controlling elements: maize. Components that may be identified and characterized; serve to modify gene action and at particular times and in particular manners.

6. The effects of hormones: act at gene level. Effects only in certain cells. Combine with chromosome - some evidence of this

III. The components of the chromosomes in nucleated organisms:

1. Enormous difference in associated components of DNA between bacteria and organisms with chromosomes and nuclei.

2. This difference: undoubtedly associated with mechanisms of reproduction of the chromosome and with control of gene action during differentiation and in individual cells, already differentiat

3. Number of DNA molecules per chromosome: few. Number of replicons: a number of them per chromosome.

4. The components of the chromosome:

   Histones: Lycine rich and arginine rich:

   Residual protein: acidic

   The new RNA species: with histones; with basic protein.

   The phosphoproteins; the phospholipids.

5. The Histones: have been the candidate for repression of gene action:

   a). Histones in active and inactive chromosome parts: not different. Some turn-over of histones without replication of DNA.

   b). What histones might be doing: describe shortly;

6. The organization of parts of chromosomes in the light microscope:

   (1). The nucleolus organizer: function; cytoplasmic ribosomes.
   Slides 1, 2. Relation to beetle Chr. SE ——— B. subtilis

   (2). The true heterochromatin in the chromosomes: psotions.
   About centromeres in Drosophila and other organisms. Appearance in the nucleus.

   At ends of chromosomes; At special regions: knobs in maize.

   (3). The position of the different parts in the working nucleus:

   Slides 3, 4, and 5.

   (4). THE IMPORTANCE OF THE RELATION OF PARTS OF CHROMOSOMES TO THE NUCLEAR MEMBRANE!

IV. The activation and repression of genes through differential condensations:

TWO CLASSES OF DIFFERENTIAL CONDENSATION: EACH AFFECTS REPRESSION OF GENE ACTION.

I. CLASS I: Chr. ———————————————

   (1). Relation of condensation to gene action: long known by cytologist: Example in two nucleated pollen grain.

   (2). Examples of differential condensation in nuclei of cells of same tissue: Slides 6 and 7.

(3). The extensi e studies of Calf thymus lymphocytes in Mirsky laboratory, Rockefeller Institute.

a). The appearance of the nuclei: Before and after swelling. Slide 8. Slide 9.

b). The position of the condensed parts with respect to the nuclear membrane.

c). The tests of the active and the inactive chromatin: Radioactive uracil - positions of formation of RNA Autoradiograph: Slide 10. (position of c ndensed parts).

d). The mode of connections of the parts in the condensed regi n:

Cross linkages betw en strands produced by lycine rich histones: associated with phosphoric acid group of DNA

This histone associated with the repression process through maintaining the clumps.

Arginine rich histones: also associated with the phosphoric acid group of DNA but combine along side of DNA.

(4). Each nucleus, has its own type of contraction of chromatin, involving diffe ent parts of the chromosomes-- diffe ent genes.

(5). The major question: how does the differential control take place? What genetic components are involved in this?

(6). A schedule of differential sequences of condensations must be present from zygote stage on, if condensation is one of the mechanisms of control of gene action.

(7). Are there special elements in the chromosmes that are associated with this? Return to case of control of esterase alleles in maize: control of time of action is different for each allele. The control of this for these alleles is associated with some component at the locus of the gene itself.

Controlling elements in maize: these are candidates for such elements that serve as co trols of gene action:

(8). Do we h ve other evidence of controlling elements or c ntrol regions within the chromosome?

(a). The behavior of the B-type chromosome in maize at pollen division. Control of non-disjunction:

The loc tion of the signaler for this: at heterochr m tic end.
The location of the ~~receiver~~ to the signals: near the centromere end of the chro osome.

Both must be present for non-disjunct on to occur.

Evidence from rye chromosome: B-tye here: similar to maize Two element system of co trol of non-disjunctions, at specific stage in development.

(b) The relationship of stage to the effectiveness of controlling elements. This brings us to the Class II type of condensation.

CLASS II CONDENSATIONS: Condensation of consequtive regions:

Parts of a chromosome; whole chromosome; whole set of chrs.
Known to be inactive for genes. IMPORTANT

1. Example: X chromosome in mammals:

XY, XX, XXX, XXXX, XXXX: Selection of X to be non-contracted, if more than one present. all others are contracted.

Position of X in nucleus: Slide 11

Hypotheses: All X chromosomes conditioned for contraction; one only is rescued from this.

2. The control region for the contraction: the responder to some signal. In one region of X chromosome.

The tests in mouse: X Autosome translocations.

Results: contraction to both sides of the control region including autosome part

3. Condensation of whole set of chromosomes: mealy bugs: males the set received from the father: Slide 12. Position in nucleus.

This set represents the set previously received from the grandmother.

Outline: Females: Female producing;

Same female, eggs laid later: Male producing.

Set from father in females: euchromatic

Set from father in males: heterochromatic.

IMPORTANT: Set from father: conditioned to condense; rescue from this occurs in eggs destined to produce females; no rescue from this if it is to produce a male.

SETTING PROCESS: Setting occurs in the germline of the father in the chromosome set it had received from its mother.

Heterochromatic set it received from its father is discarded; does not get into sperm.

4. The relation between setting for heterochromatization and for elimination: uses the same mechanism: Evolutionary sequence

Some mealy bugs: Set from father is eliminated in early cleavage of cells destined to become soma cells. This important for my thesis.

V. The control of the elimination process: reaction of chromosomes destined for elimination during cleavage to cytoplasmic substance which rescues this: Cecidomyid.

1. The Egg: Pole plasm. The reticulate substance in pole plasm. The pole plasm and the germ line.

a). Normal behavior:

b). To show the rescue from elimination related to reticulate substance (but not the pole plasm). Example of one type of test:

Ligature:

c). Centrifugation studies: Any nucleus that comes adjacent to reticulate substance will have its E chromosomes rescued from elimination process.

VI. The tests of the responder and the signaler for the elimination process in Sciara and the rescue mechanism. Its relation to control of condensation of only one X in mammals: the rescue from condensation of only one X.

1. Sciara germ line of male: later stages; Meiosis; constitution of sperm.

2. The Females: Two types:

X' X: Produces only females, normally

X X: Produces males only.

To show that the signaler is, in the X' and not the X chromosome and that the receiver is in the heterochromatin, at one location in the X, received from the father. This receiver component was set in male germ line: to effect elimination of both X chromosomes during 7th or 8th division.

The eliminations of X in soma of eggs produced by
Eggs of X' X females: only one of two sister X chromosomes from father
" " X X Females: both X chromosomes from father eliminated.

3. Relation of control of elimination in the soma cells to product of the X' chromosome:

Females: X' X== normall female produces: one X from male eliminated.

Non-disjunctions:at meiosis: No X chromosome in egg nucleus

Zygote: 1 A + 2 X 1 A

Soma elimination: One X only eliminated.

Female: XX= normal zygote 1X + 1A 2X + 1A.
Soma: elimination of both X from father.

Nondisjunctions: 2 X chromosomes in egg from mother.

Zygote: 2 X + 1 A / 2 X + 1 A.

Both X chromosomes from male eliminated.

Hypothesis: Both X chromosomes from father destined for elimination in the soma. X chromosome produces some "particle" that is able to rescue only <u>one</u> X chromosome. Must be in cytoplasm as in non-disjunction case, no X from female in the zygotes.

Setting: Occurs to element located in heterochromatin of X:

<u>XXXX</u> ◊———————————

X- Autosome translocations:

Any part of chromosome complement that carries this element will follow elimination path, non-disjunction path at meiosis of male.

Best translocation: <u>XXX</u>.....◊.....

.....◊ X———————————

Setting occurs during germ line of male to this element.

Rescue occurs in soma of females to one element

No settings occur to this element, leading to elimination in germ line of female. Once rescued, remaines rescued until it again passes through germ line of male.

4. The relation of the Sciara case to X chromosomes in mammals: The setting region:for condensation.

Only one X rescued. Signaler not in X chromosome probably. Should be in one of the autosomes.

VII. The setting of the controlling elements in maize and the resetting process. Will discuss later.