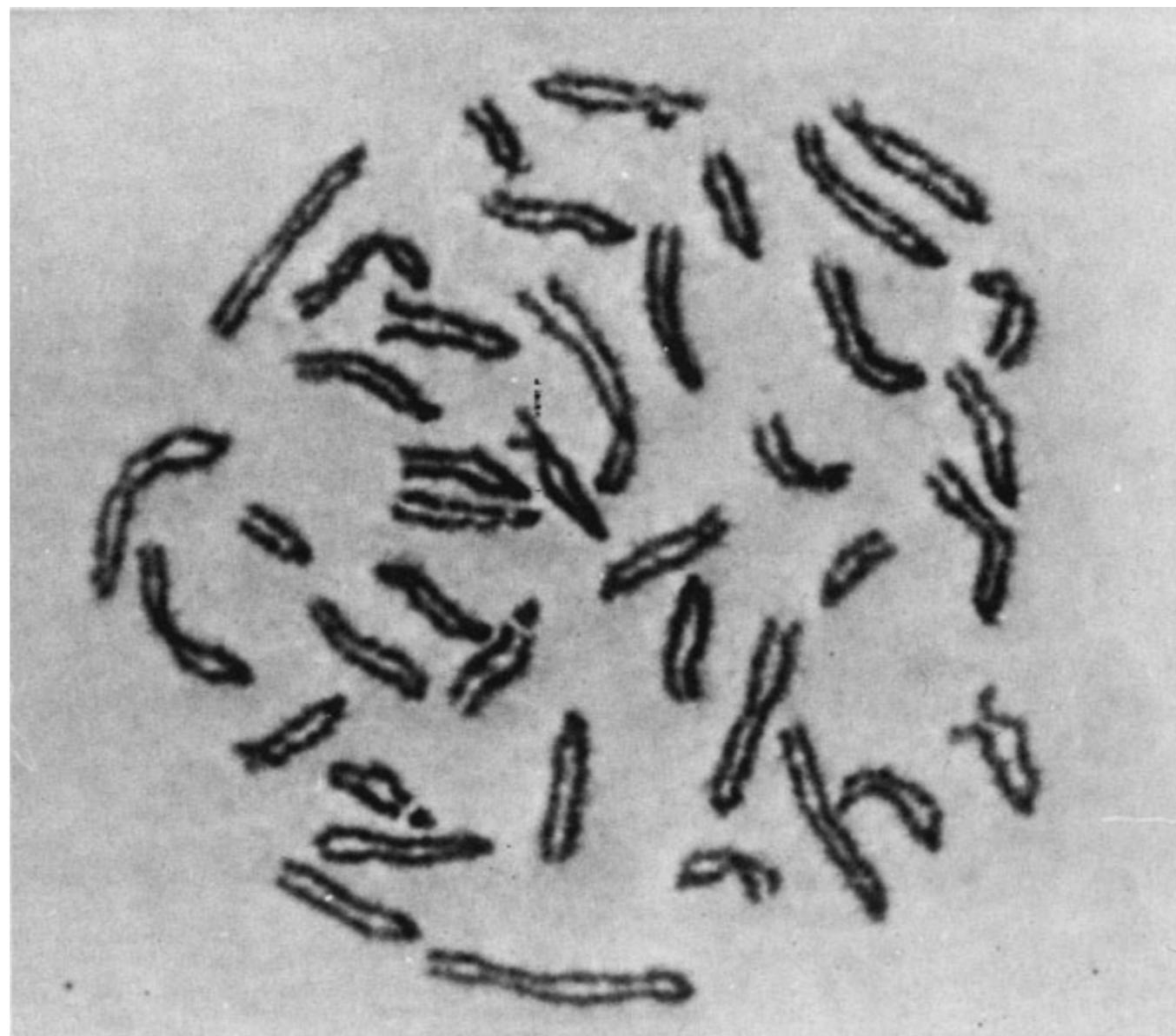
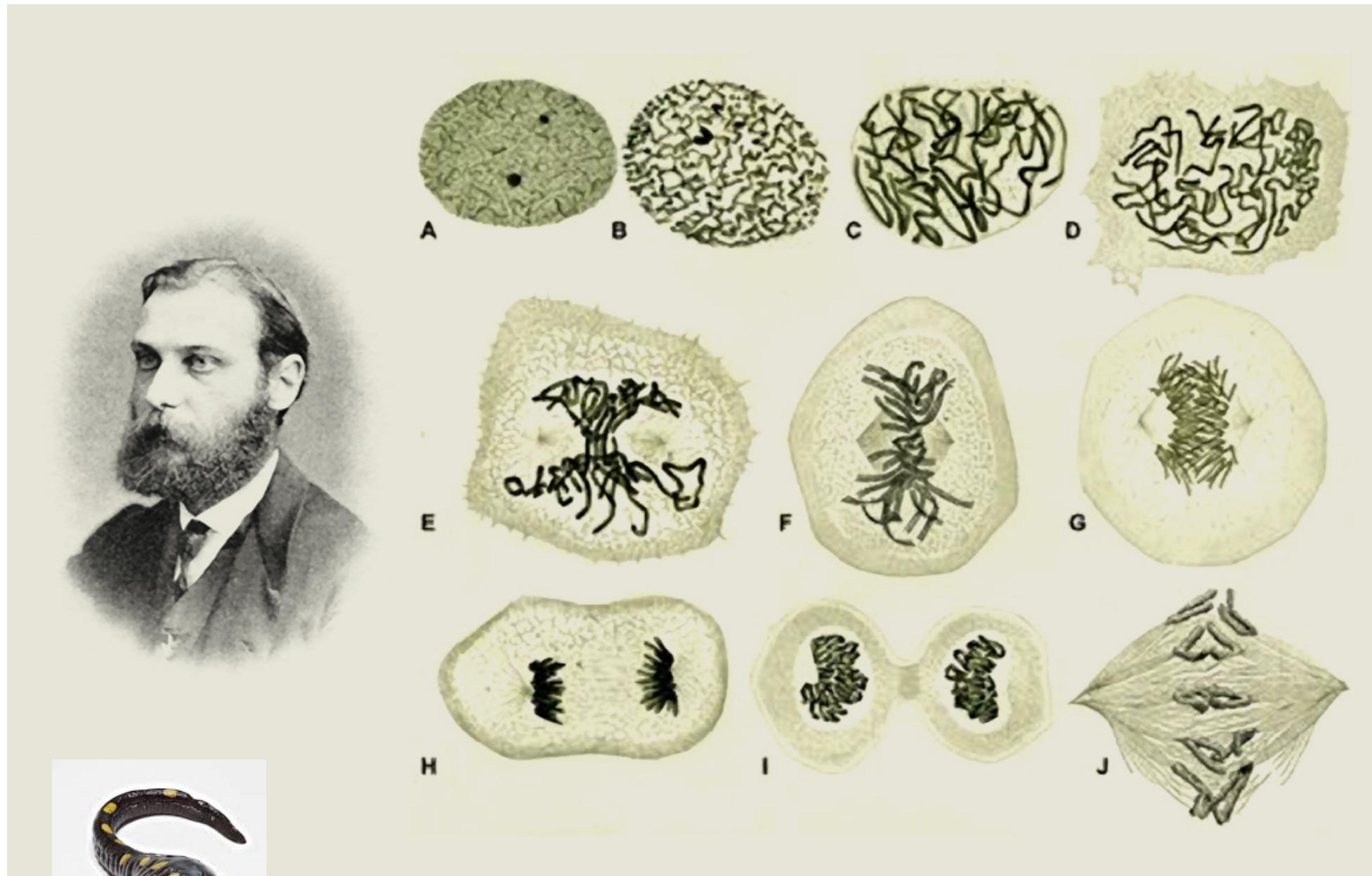


# Bio393: Genetic Analysis

Chromosome theory, recombination, and mapping



# Walther Flemming stained cells

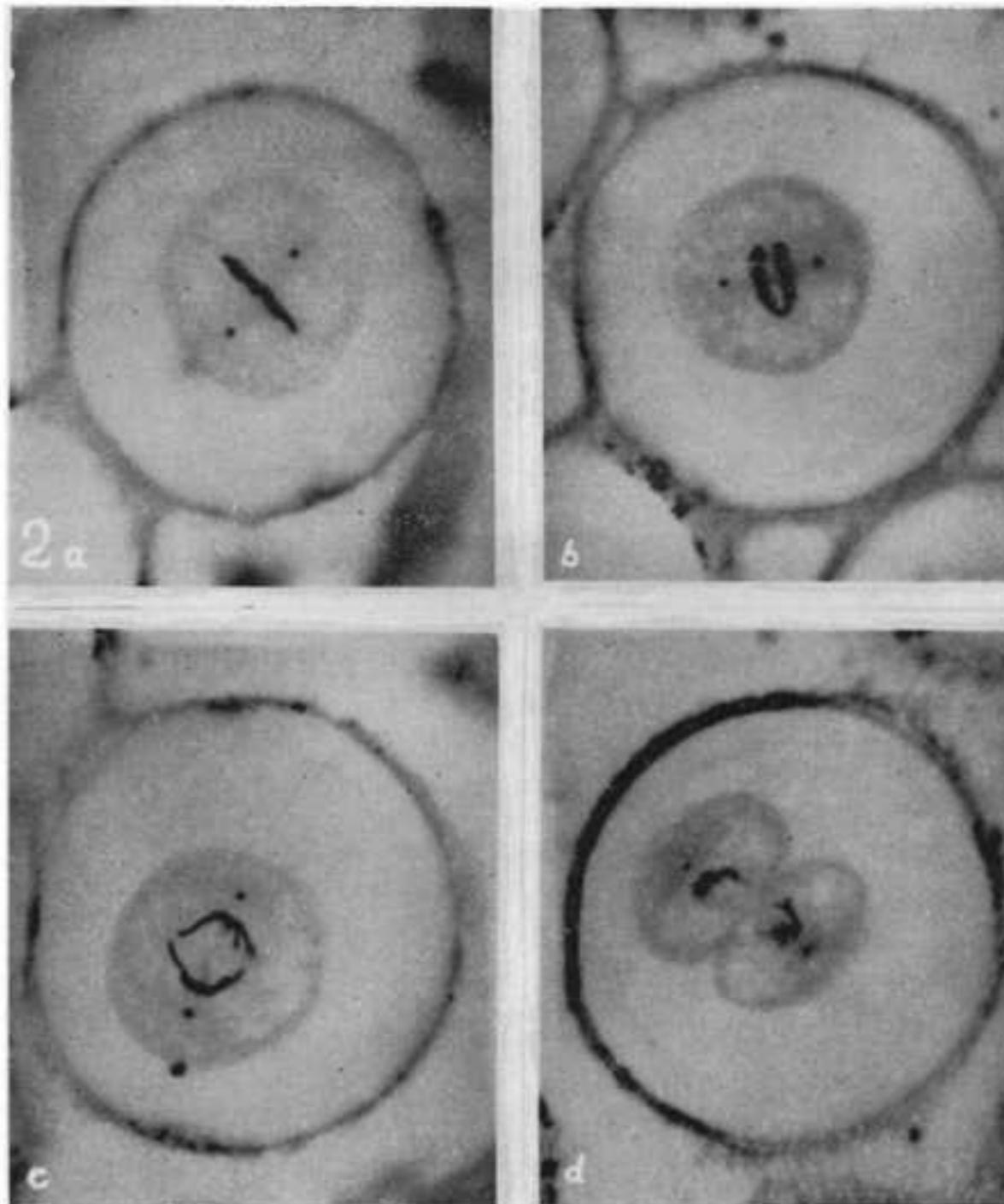


Walther Flemming, 1882

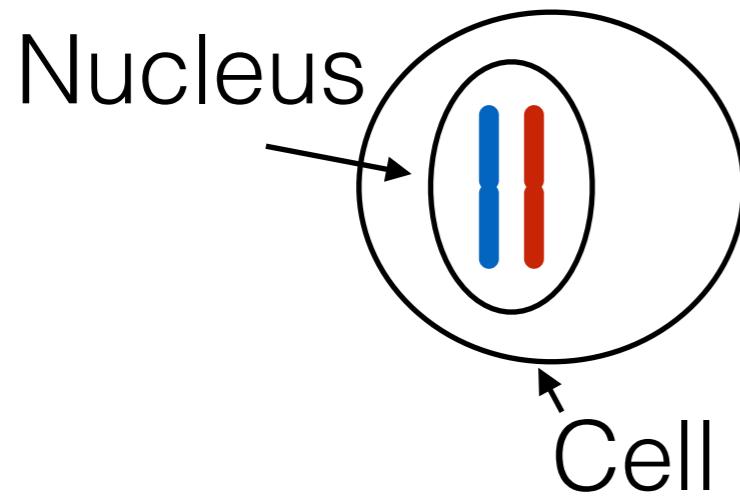
# Cells divide their chromosomes with high fidelity



Theodor Boveri



# Terms for mitosis and meiosis



- | Chromosome
- || Pair of homologs (2N)
- X Sister chromatids

Ploidy (N)  
Diploid (2N)  
Haploid (1N)  
Polyploid (>2N)  
Gamete

# Gametes have half the chromosomes of the soma



Theodor Boveri



*Parascaris equorum*

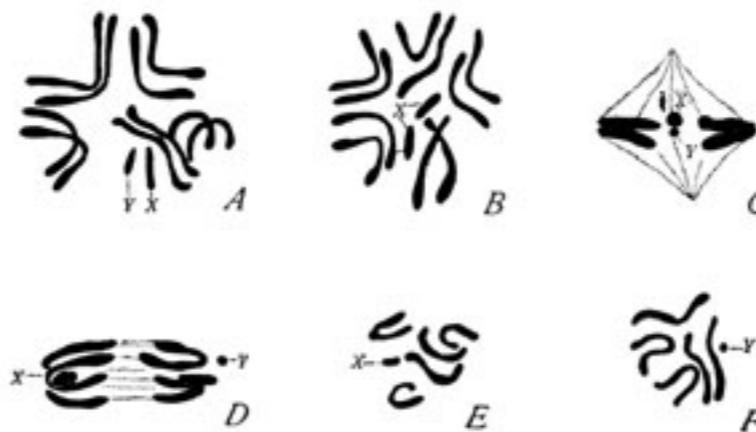


# Discovery of sex chromosomes



Courtesy of the Marine Biological Laboratory.  
Noncommercial, educational use only.

Nettie Stevens



*Tenebrio melitor*

then 50 beetle species and nine species of fly!

Lecture 2

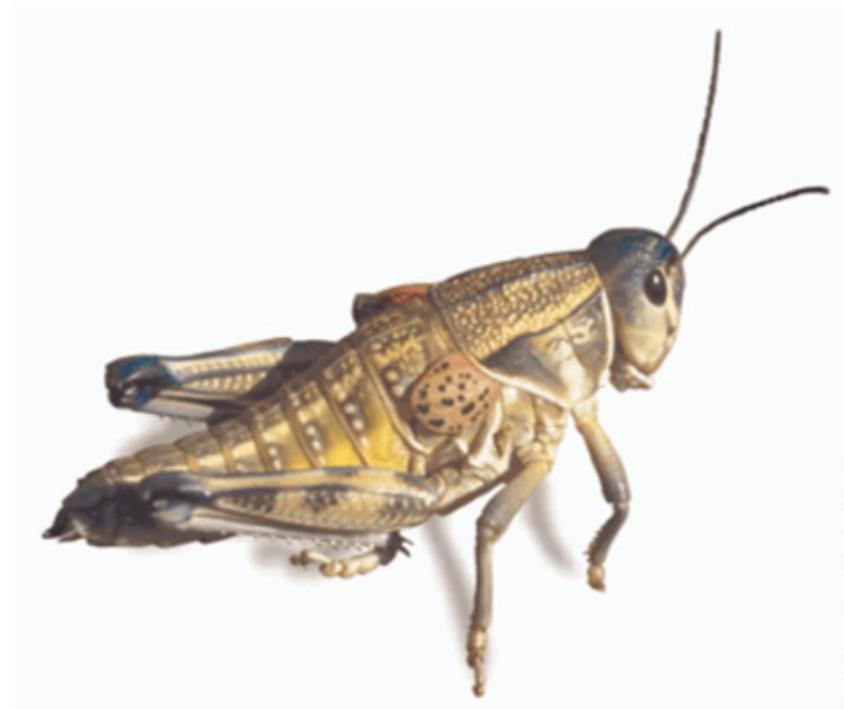
# Discovery of a connection to Mendel's principles



Walter Sutton



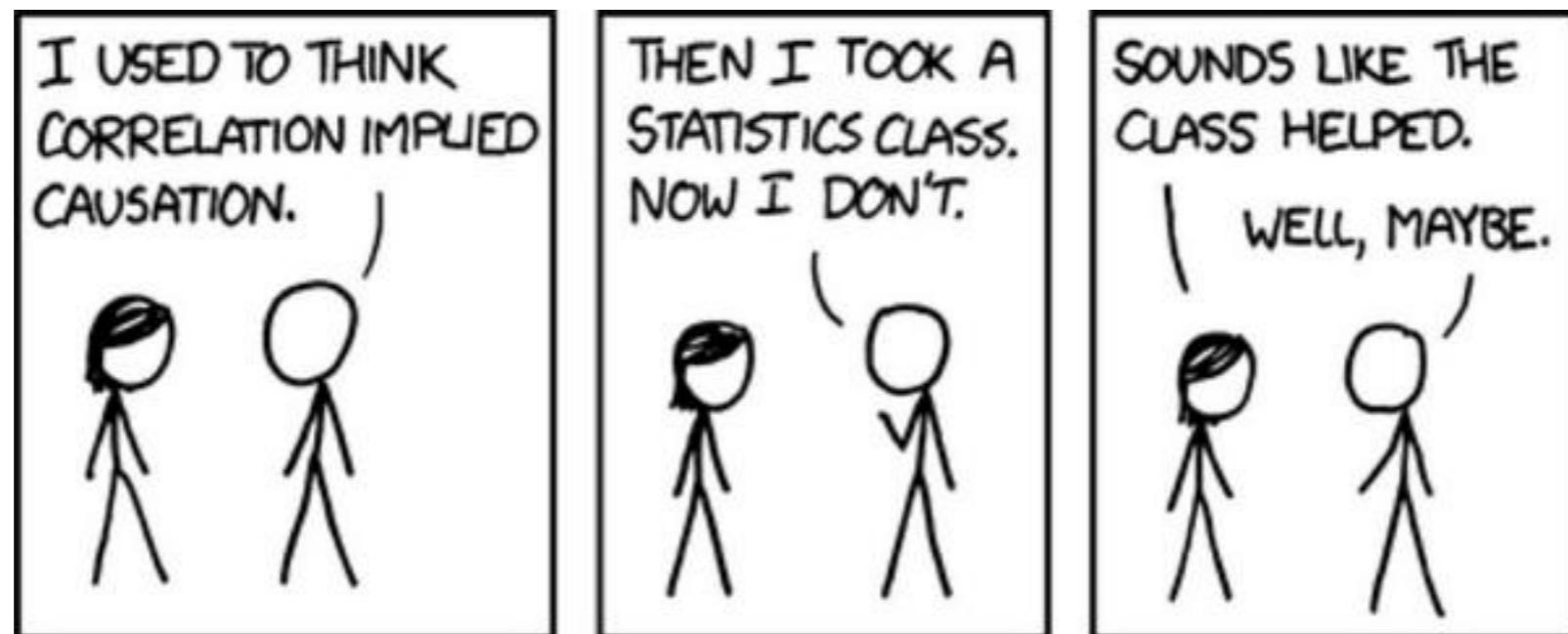
Michael Abbey/Photo Researchers, Inc.



E.R. Deggingen/Color-Pic, Inc.

- Gametes have half chromosome complement of somatic cells
- Homolog separation to gamete was random

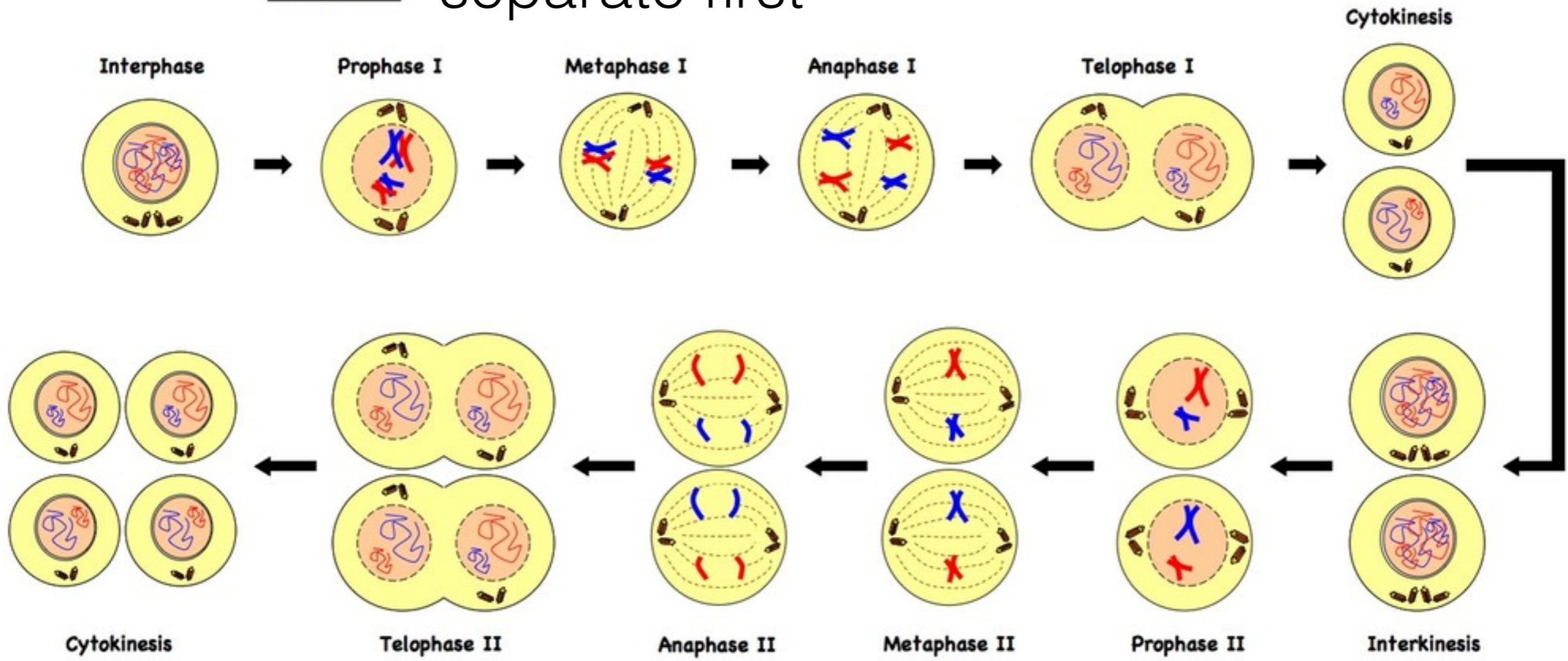
# Correlation does not mean causation



# Meiosis: A reductional division in two acts

Homologs

**MEIOSIS I** separate first



Keep track of  
centromere

Lecture 2



**Thomas Hunt Morgan**

# *Drosophila melanogaster*: genetics superstar



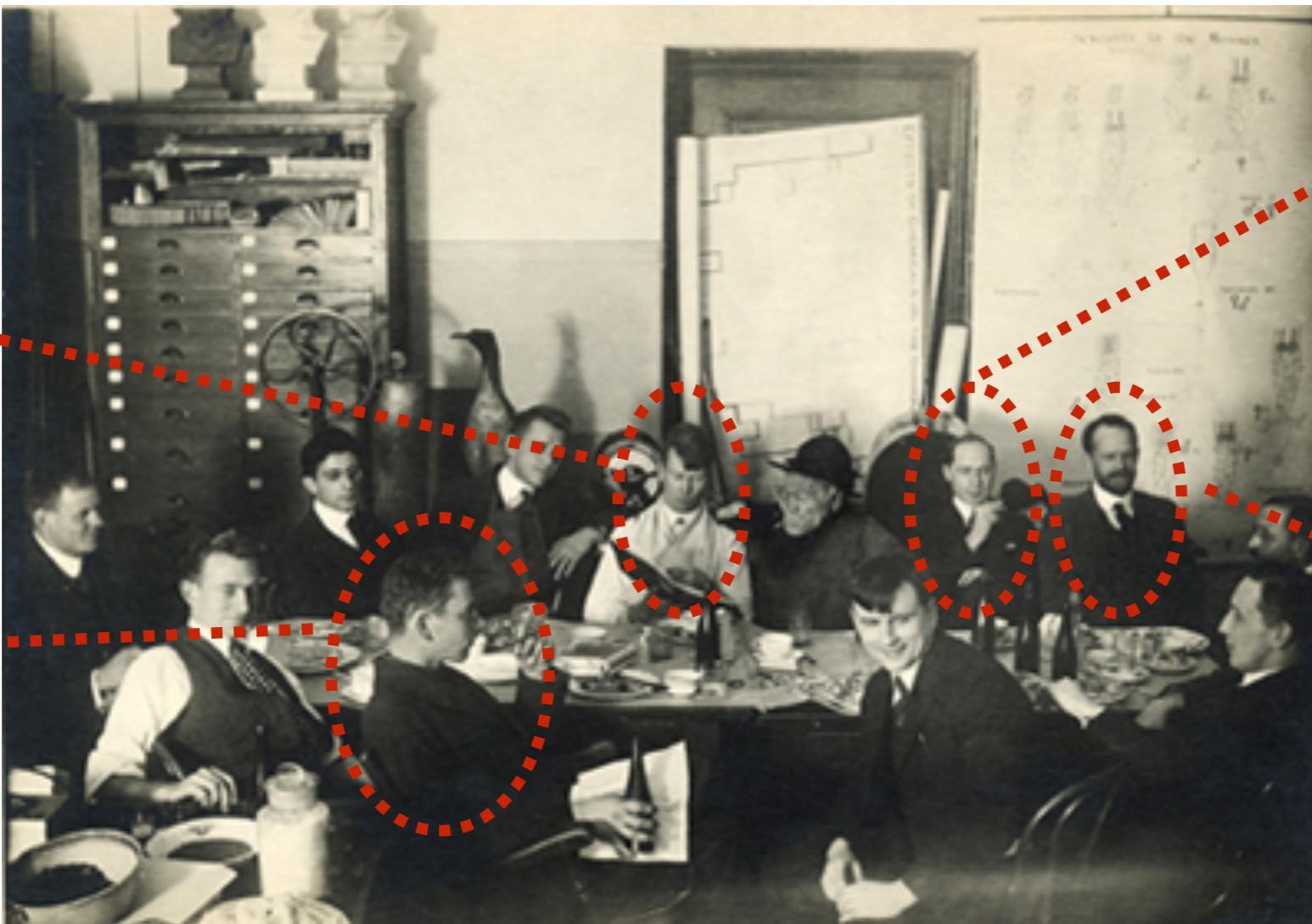
Courtesy of the Marine Biological Laboratory.  
Noncommercial, educational use only.

**Nettie Stevens**



**Thomas Hunt Morgan**

# The fly room at Columbia



Calvin Bridges



Alfred Sturtevant



Hermann Muller



Thomas H. Morgan

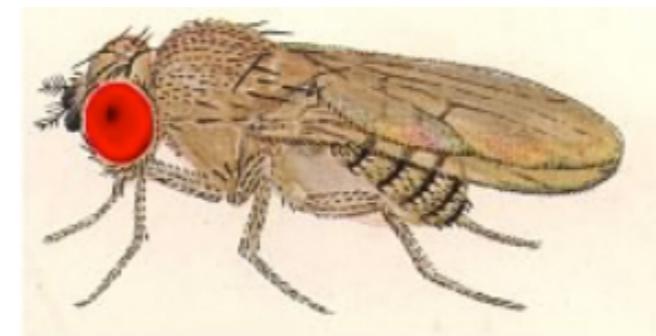
***W*<sup>+</sup>**

***W***





X



♂



♀



♂



♀

What is dominance relationship of *white* mutant allele?

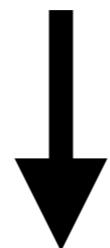


X

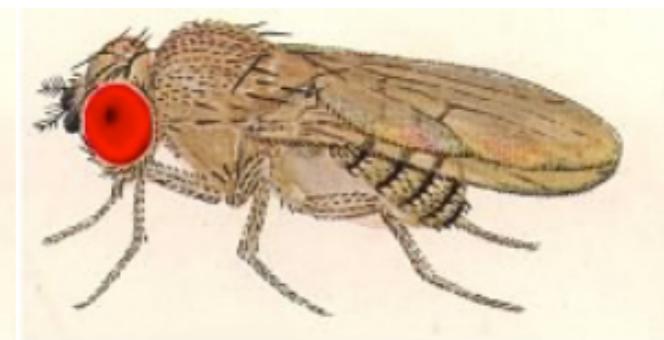
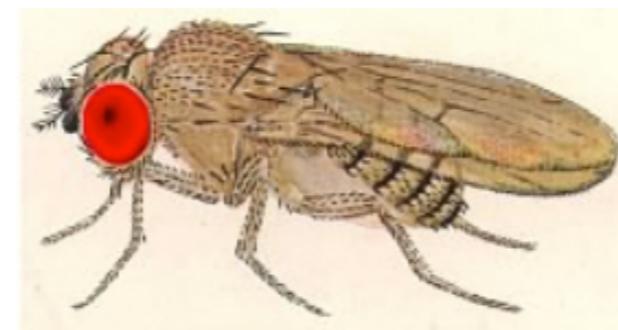


F1

♂



♀



♂

♂

♀

♀

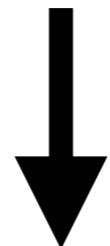
# The reciprocal cross



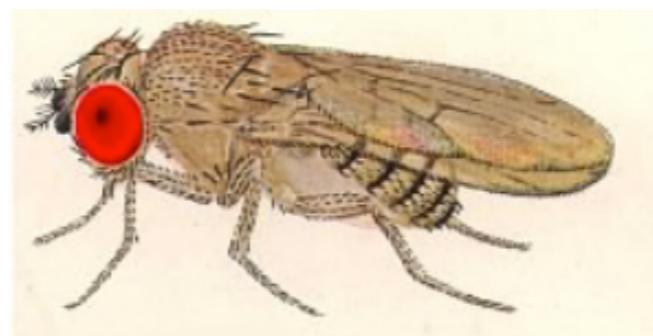
X



♂

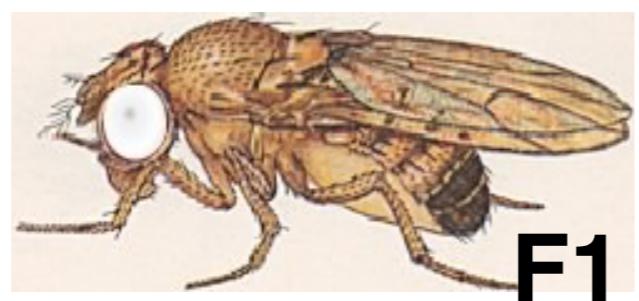


♀



♂

♀



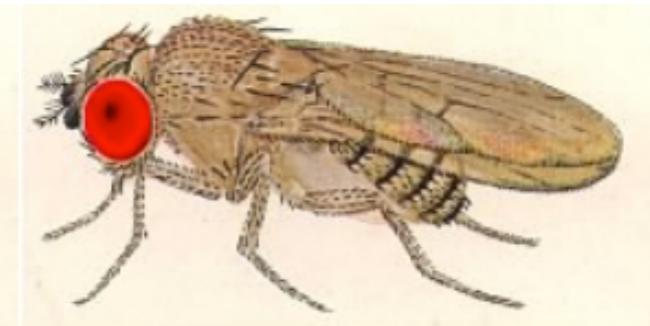
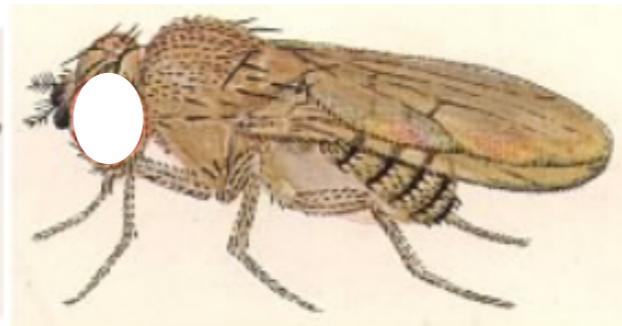
X



♂



♀



♂

♂

♀

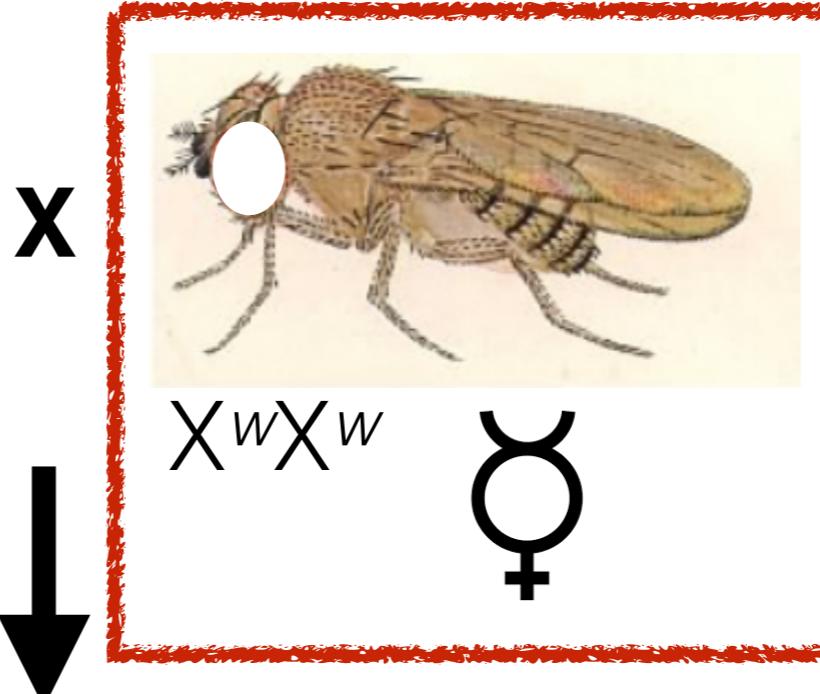
♀

Equal ratios of each sex and eye color

Lecture 2



$X^{w+}Y$  ♂



1999/2000  
offspring



$X^wY$  ♂

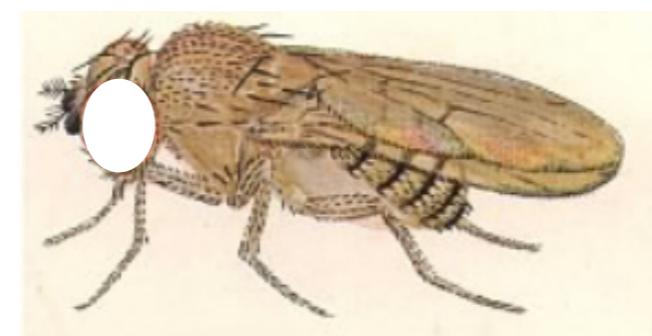


$X^wX^{w+}$  ♀

1/2000  
offspring

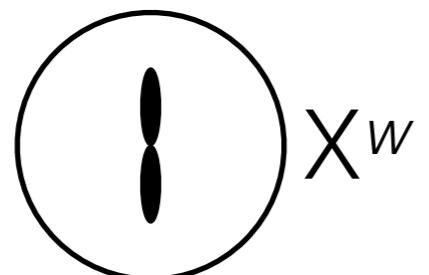
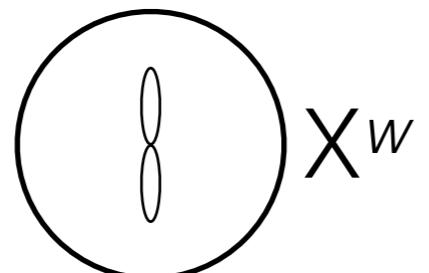
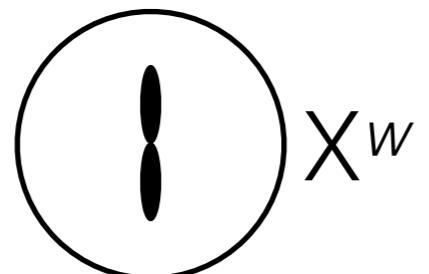
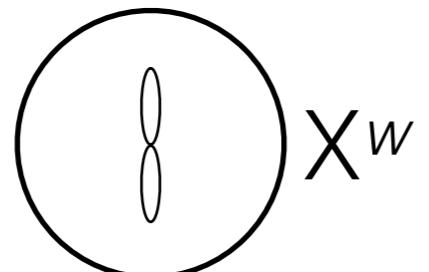


♂

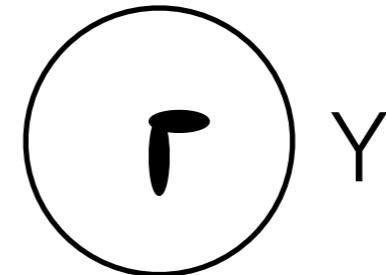
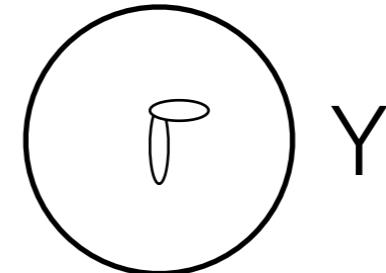
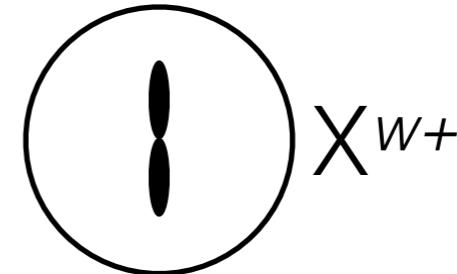
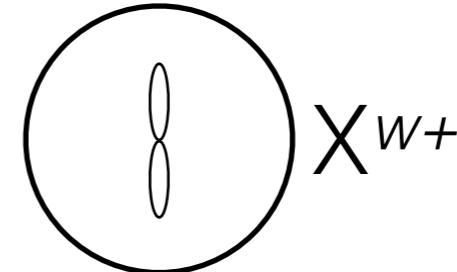


♀

Female  
gametes

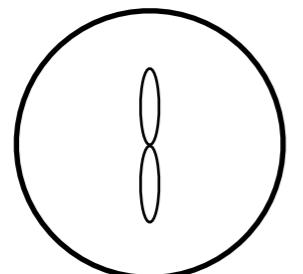
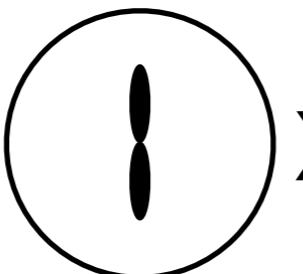
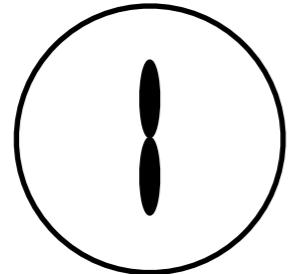
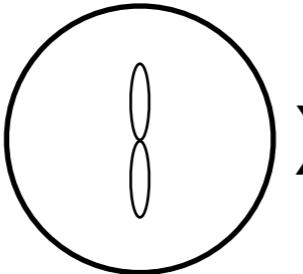
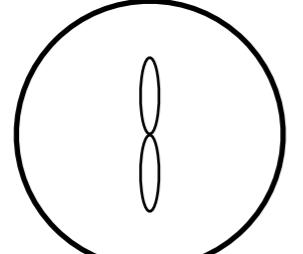
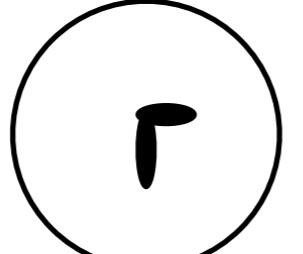
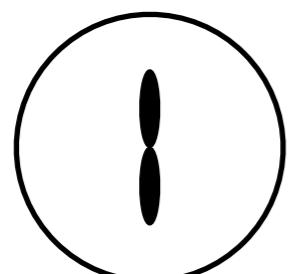
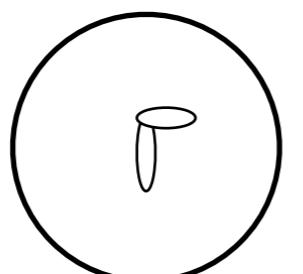
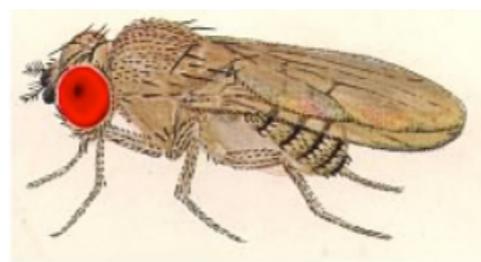


Male  
gametes



1999/2000  
offspring

Female  
gametes      Male  
gametes

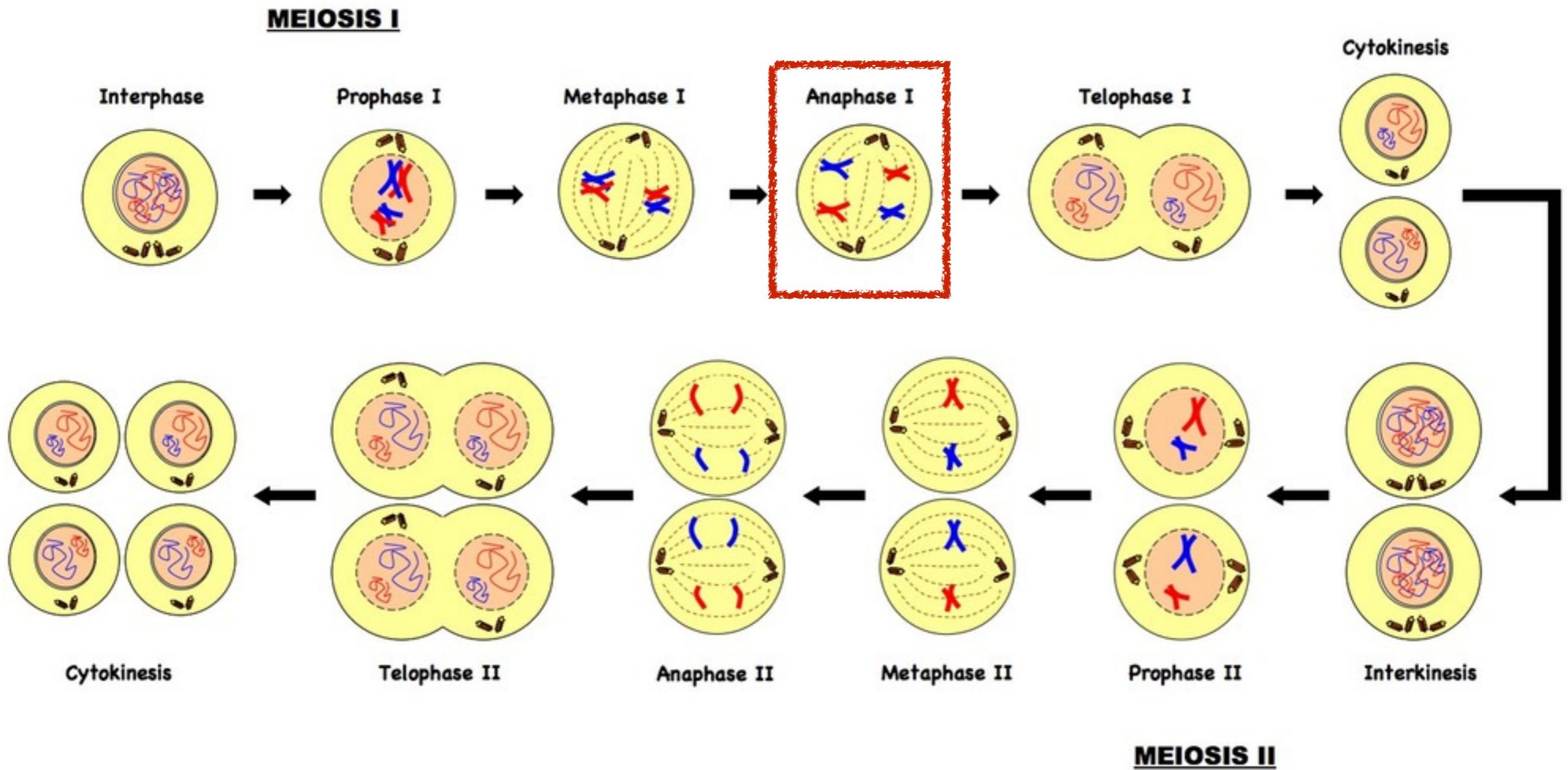
 $X^w$  $X^{w+}$  $X^w$  $X^{w+}$  $X^w$  $Y$  $X^w$  $Y$ 

Offspring

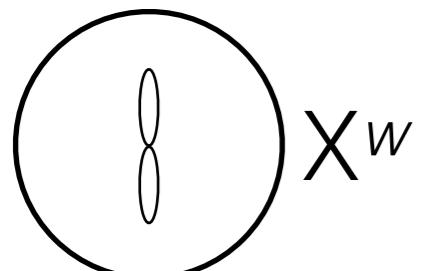
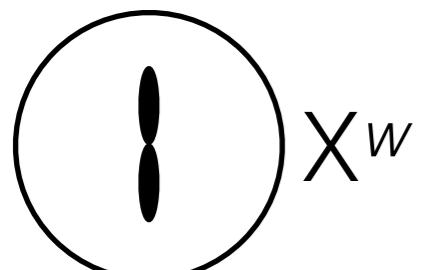
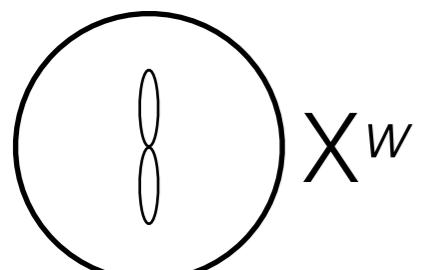
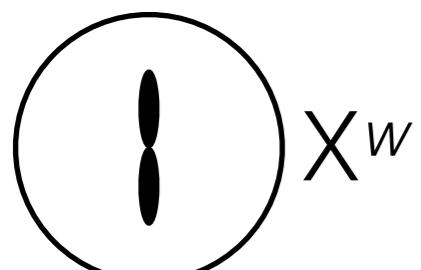
 $X^wX^{w+}$  $X^wX^{w+}$  $X^wY$  $X^wY$ 

**What is going on with the rare (1/2000) class?**

# Meiotic non-disjunction I



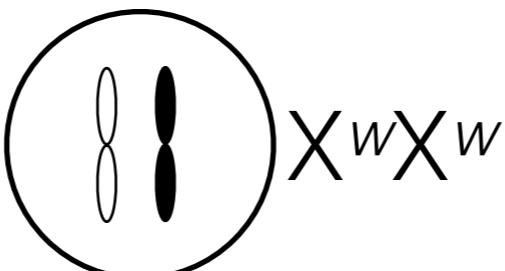
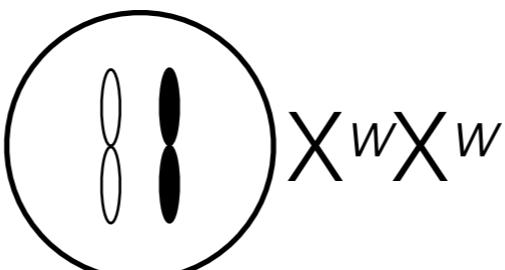
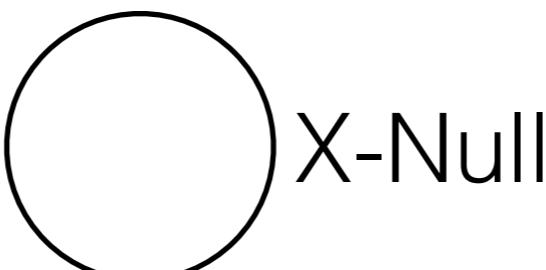
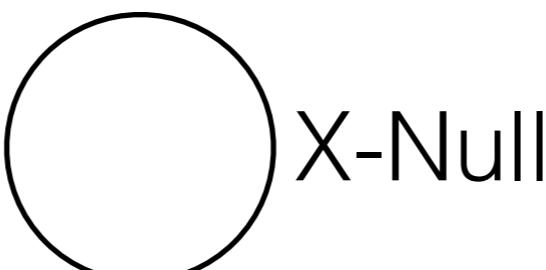
Female  
gametes

 $X^w$  $X^w$  $X^w$  $X^w$ 

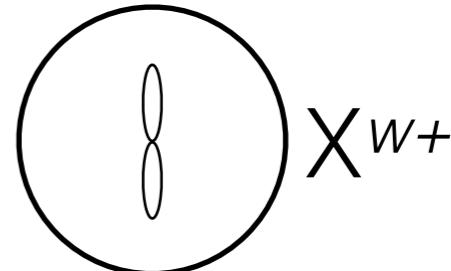
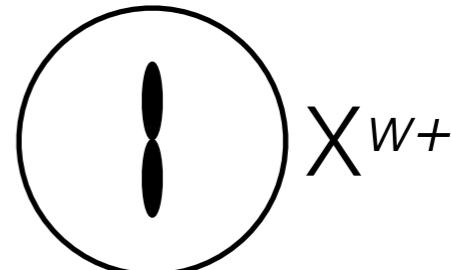
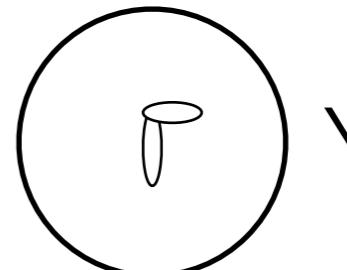
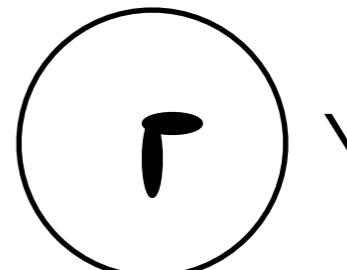
1999/2000  
offspring

 $X^{w+}X^w$  $X^wY$ 

Meiosis I NDJ  
Female gametes

 $X^wX^w$  $X^wX^w$  $X\text{-Null}$  $X\text{-Null}$ 

Male  
gametes

 $X^{w+}$  $X^{w+}$  $Y$  $Y$ 

1/2000  
offspring

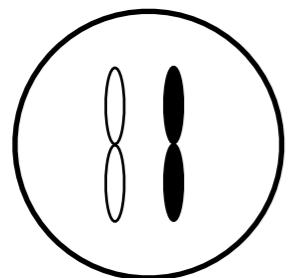
 $X^{w+}0$  $X^wX^wY$ 

red male

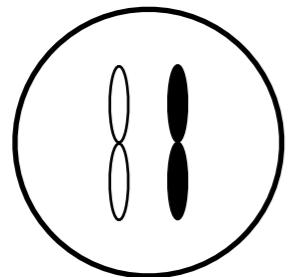
white female

**Two different types of female gametes**

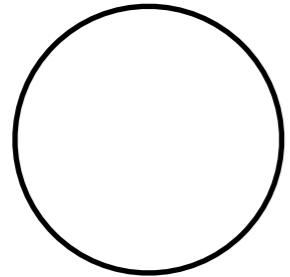
Meiosis I NDJ  
Female gametes



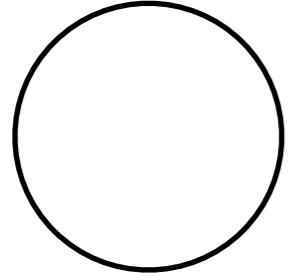
$X^wX^w$



$X^wX^+$

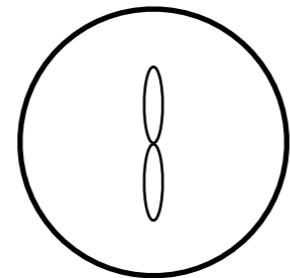


X-Null



X-Null

Male  
gametes



$X^w+$



Y



$X^w+0$



Y

Offspring

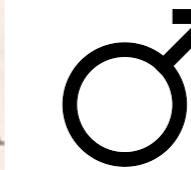


Dead

$X^wX^wX^w+$



$X^wX^wY$



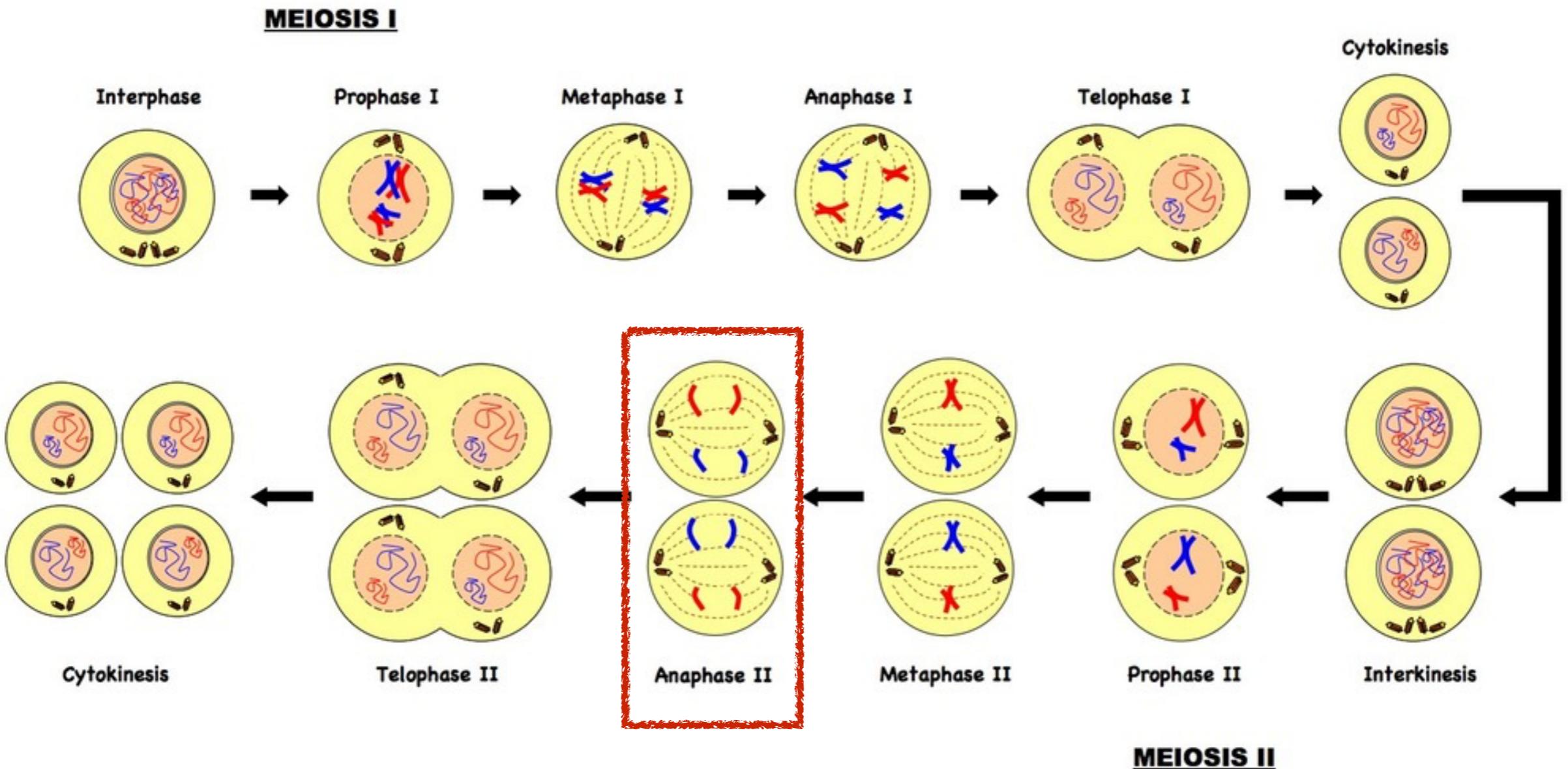
$X^w+0$



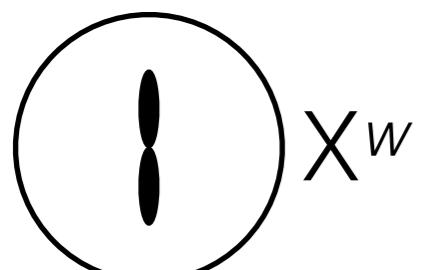
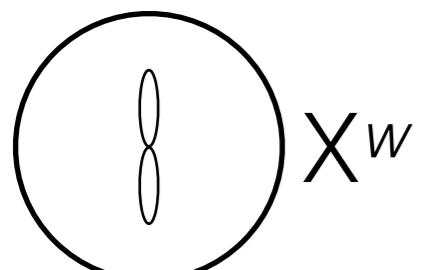
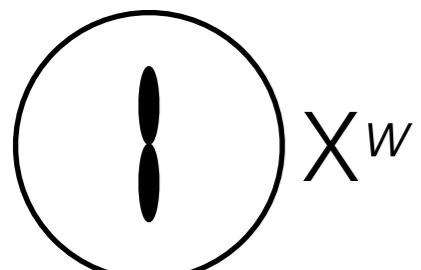
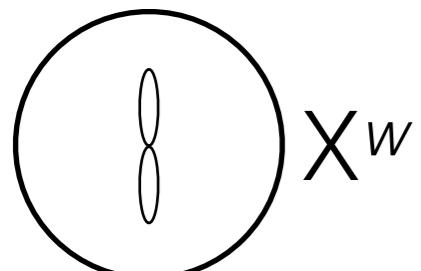
Dead

0Y

# Meiotic non-disjunction II



Female  
gametes

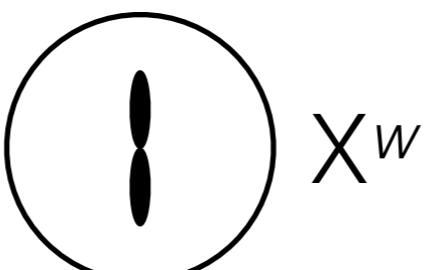
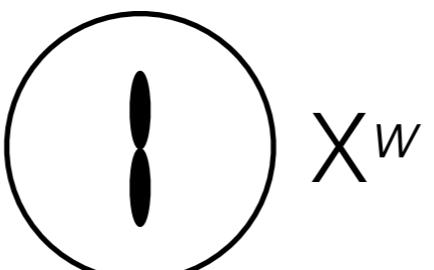
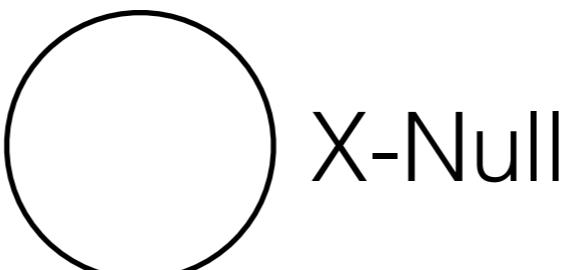


1999/2000  
offspring

$X^{w+}X^w$

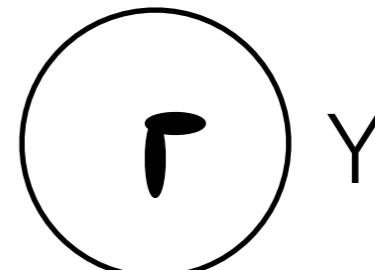
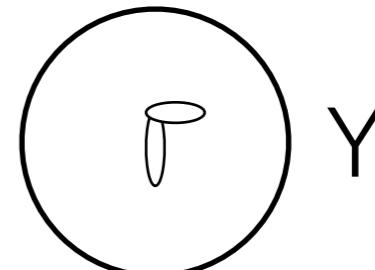
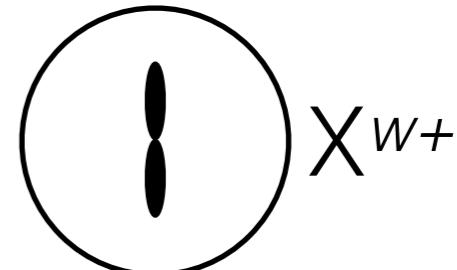
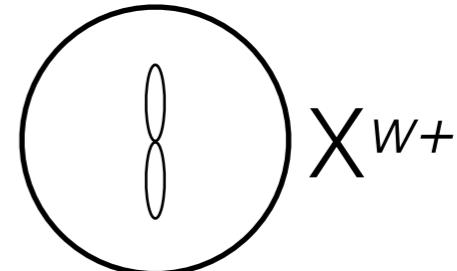
$X^wY$

Meiosis II NDJ  
Female gametes



1/2000  
offspring

Male  
gametes

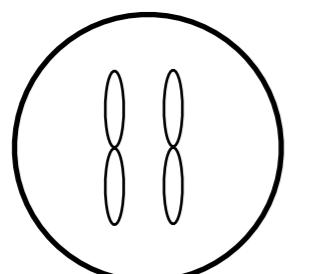


**Three different types of female gametes**

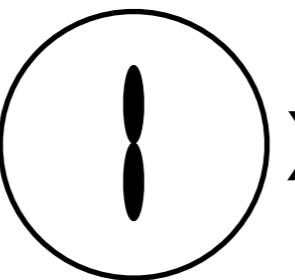
$X^{w+}X^w$        $X^{w+}0$       red male  
 $X^wY$        $X^wX^wY$       white female

Meiosis II NDJ  
Female gametes

Male  
gametes



$X^wX^w$

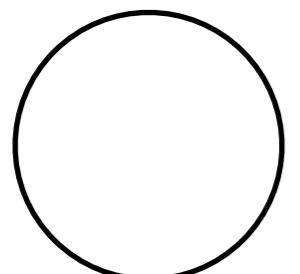


$X^{w+}$

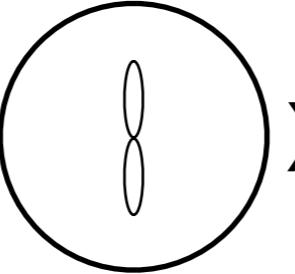


Offspring

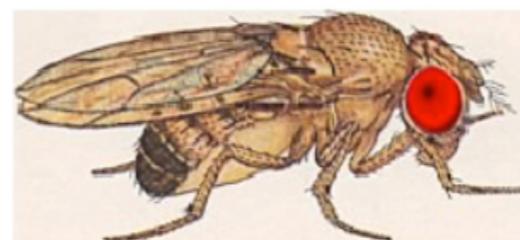
Dead  $X^wX^wX^{w+}$



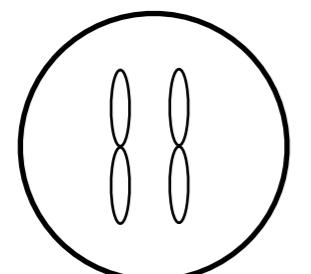
X-Null



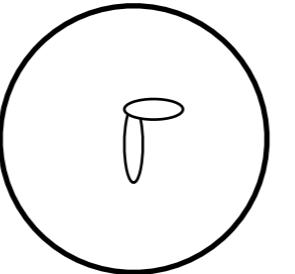
$X^{w+}$



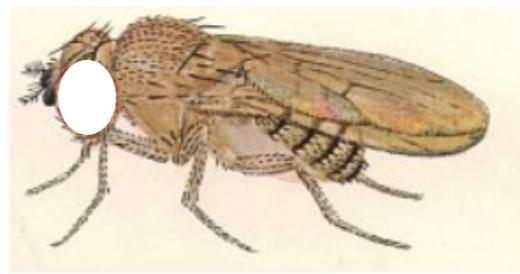
$X^{w+}0$



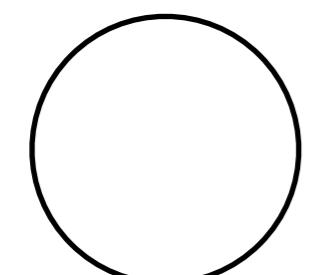
$X^wX^w$



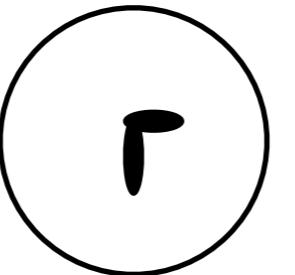
Y



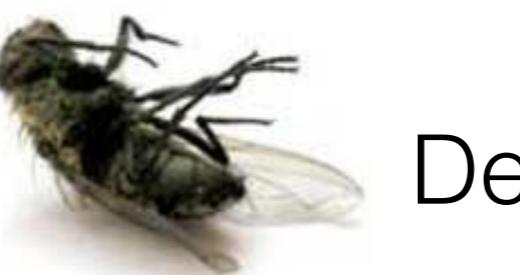
$X^wX^wY$



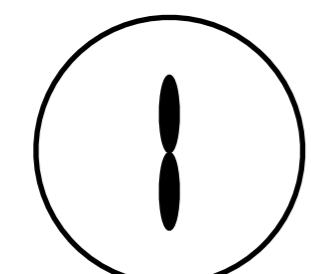
X-Null



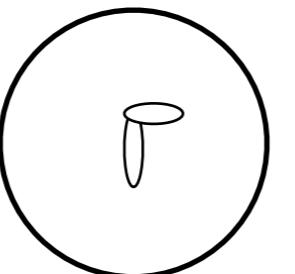
Y



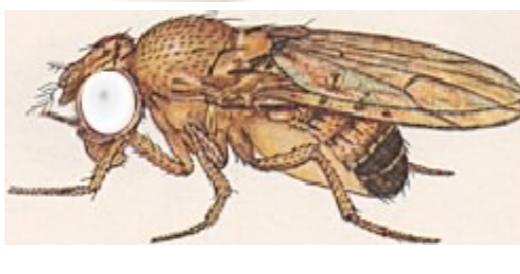
Dead 0Y



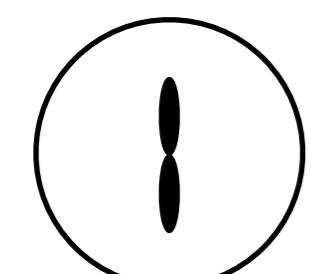
$X^w$



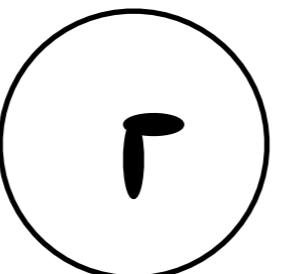
Y



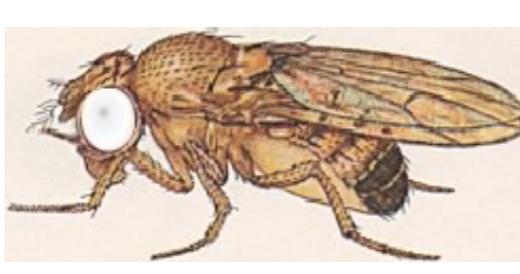
$X^wY$



$X^w$



Y



$X^wY$

# The connections between chromosome NDJ and a trait was made by Stevens and Bridges



Courtesy of the Marine Biological Laboratory.  
Noncommercial, educational use only.

**Nettie Stevens**



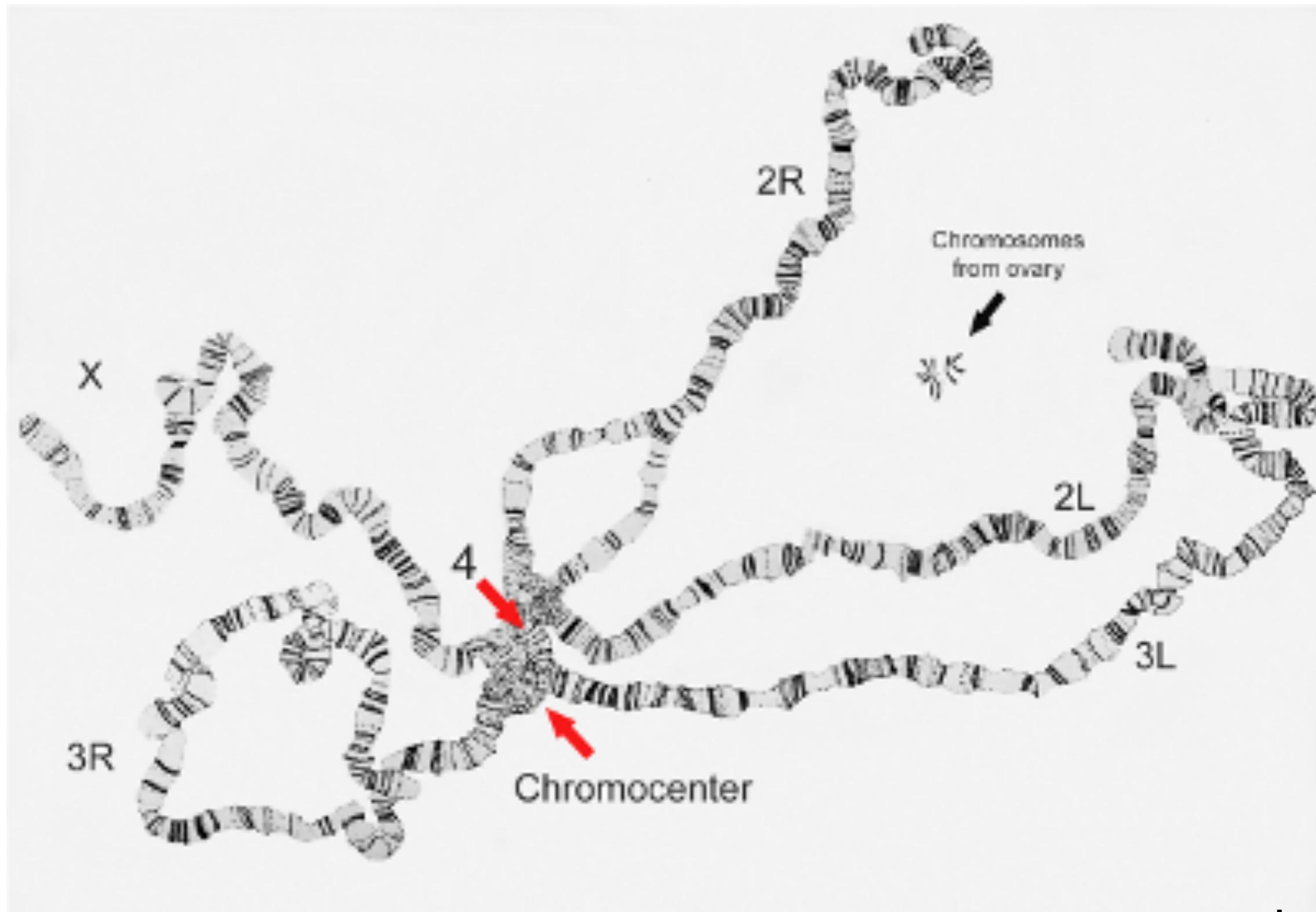
Calvin Blackman Bridges, 1927.  
Photo courtesy of Cold Spring Harbor  
Laboratory Archives.

**Calvin Bridges**



**Polytene  
chromosomes**

# *Drosophila* polytene chromosomes allow us to directly visualize genetic principles



Why did the first cross not indicate to them that something weird was going on?

$X^wY$

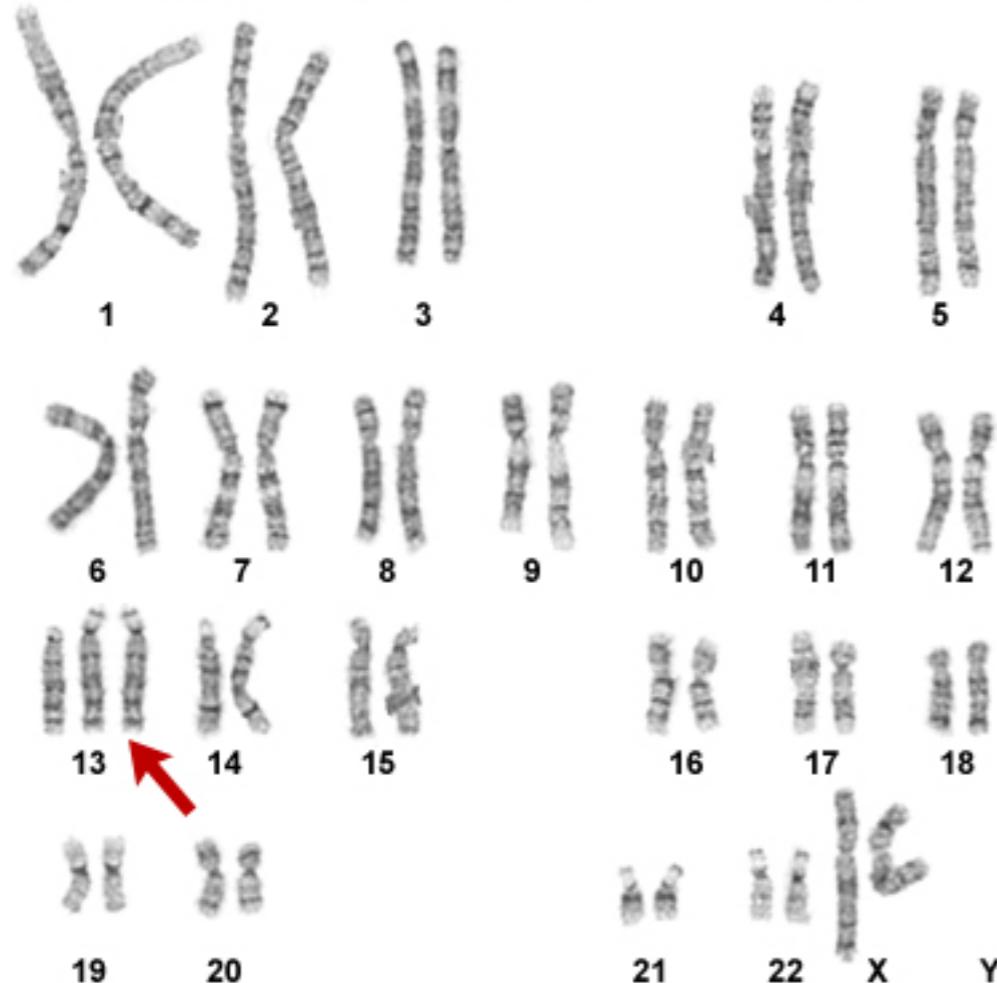
**X**

$X^{w+}X^{w+}$

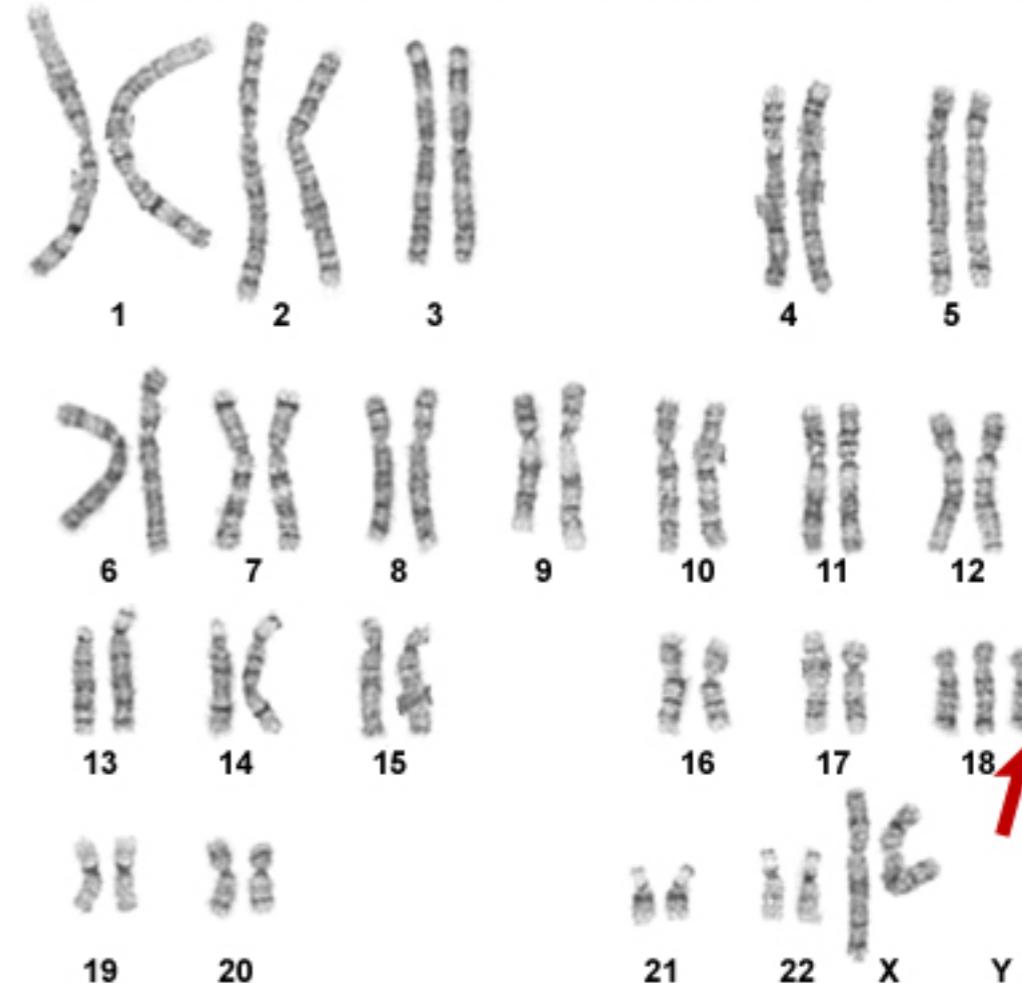
How can you tell the difference  
between Meiosis I NDJ and Meiosis II NDJ?

# Non-disjunction is a relatively common error - not just the X chromosome aneuploidy

Karyotype From a Female With Patau syndrome (47,XX,+13)



Karyotype From a Female With Edwards Syndrome (47,XX,+18)



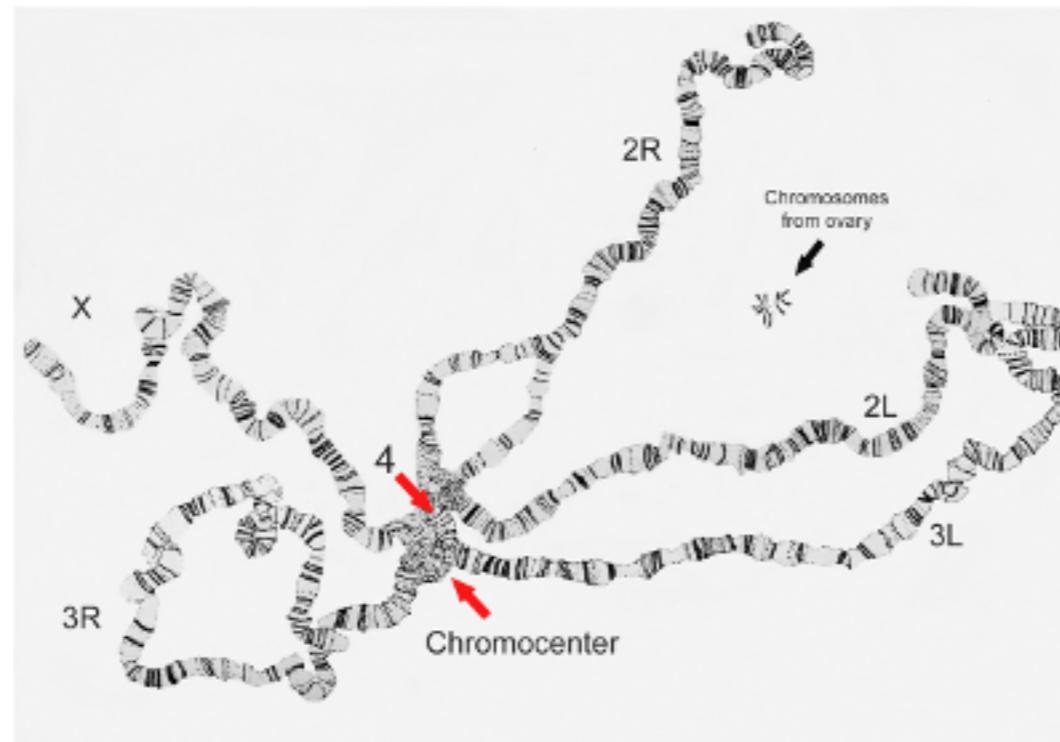
© Clinical Tools, Inc.

© Clinical Tools, Inc.

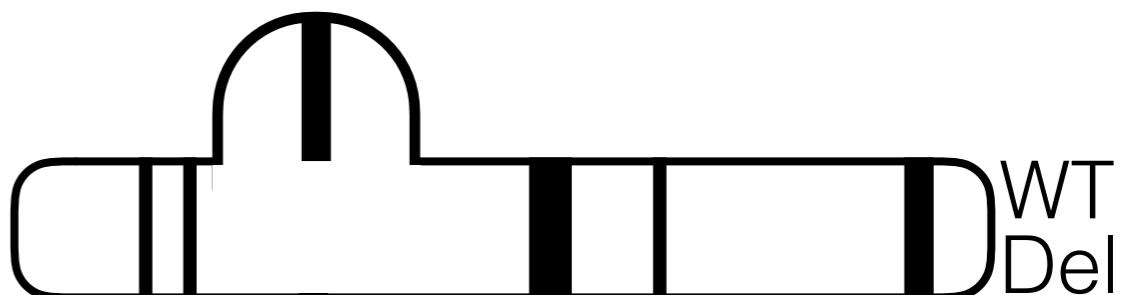
# Non-disjunction is a relatively common error - not just the X chromosome aneuploidy



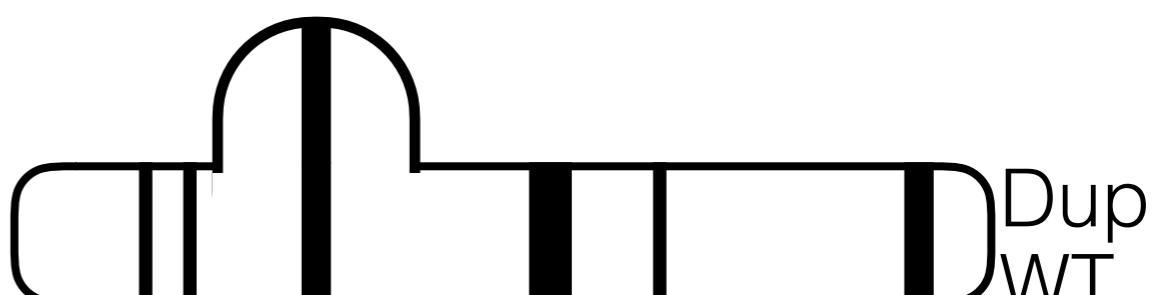
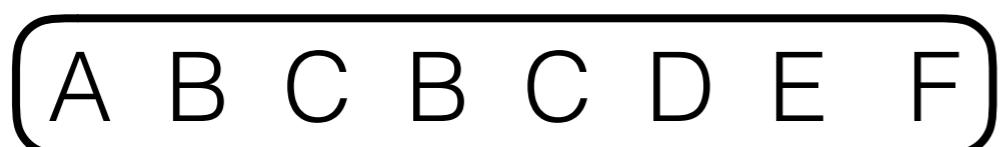
# Chromosomal abnormalities



WT



**Deletion BC**



**Duplication BC**

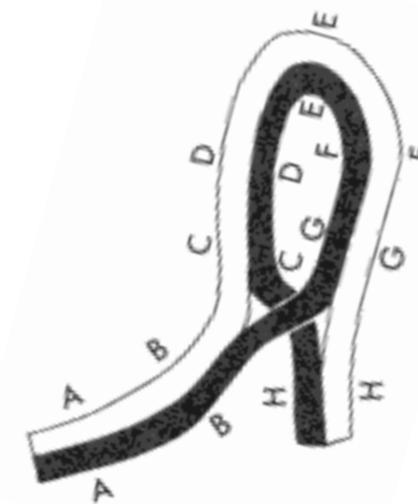
# Chromosomal abnormalities



WT



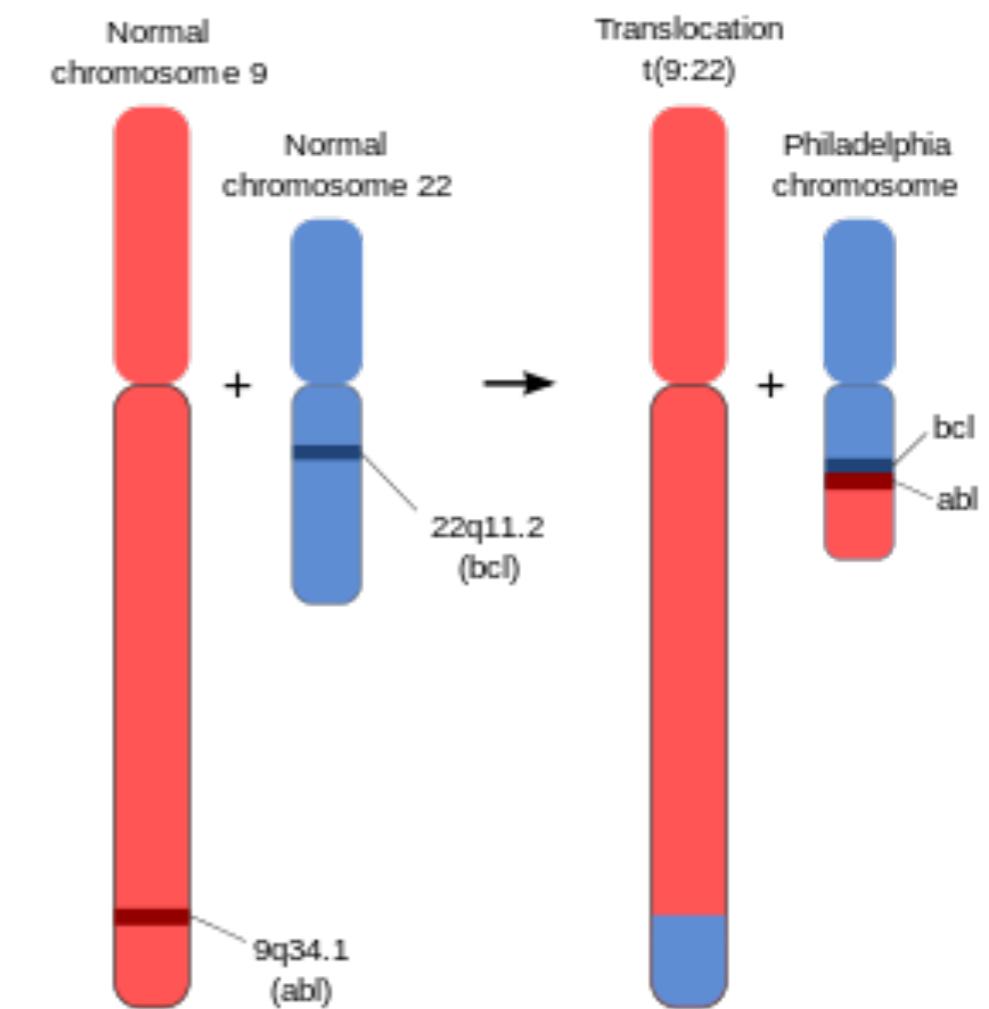
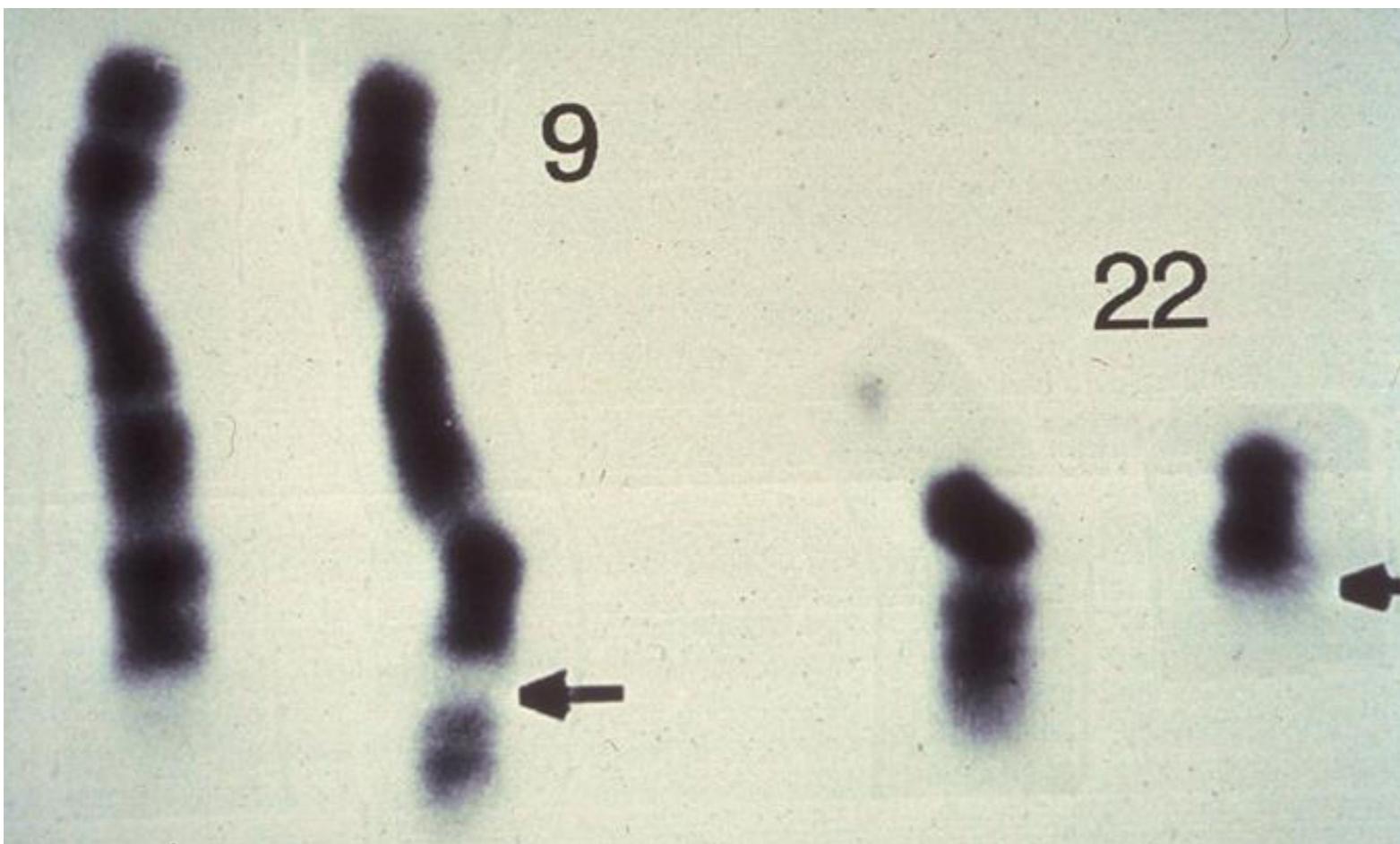
Inversion BCD



Translocation ABC-XYZ

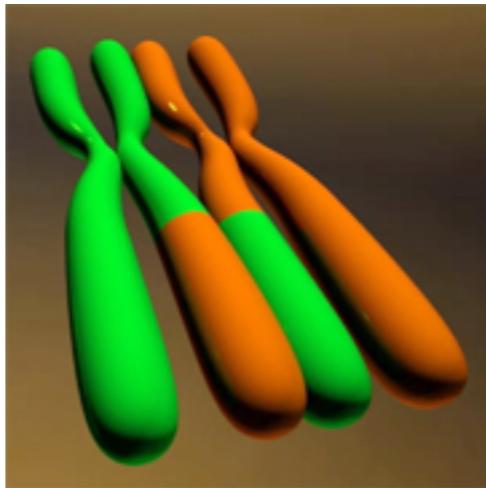
Fusion of two chromosomes

# The Philadelphia chromosome: translocation



Janet Rowley

# Recombination and mapping

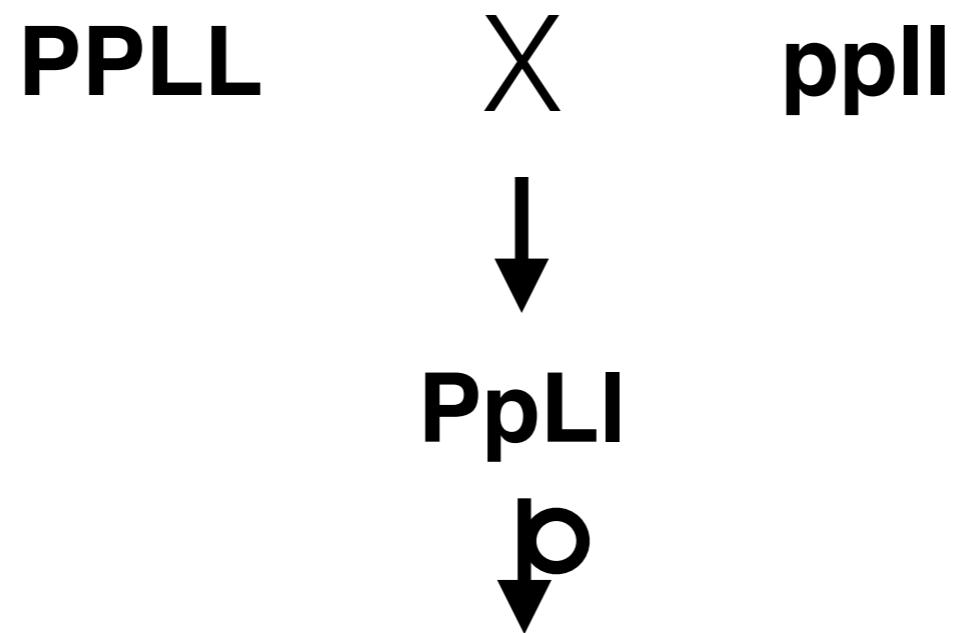




**Reginald Punnett      William Bateson**



# Bateson and Punnett's pea crosses



P= purple flower  
p= red flower  
L= long pollen  
l= short pollen

# Bateson and Punnett's pea crosses

PPLL      X      ppll



PpLl



Phenotype	Expected number	Expected ratio
Purple Long	215	9
Purple short	71	3
red Long	71	3
red short	24	1

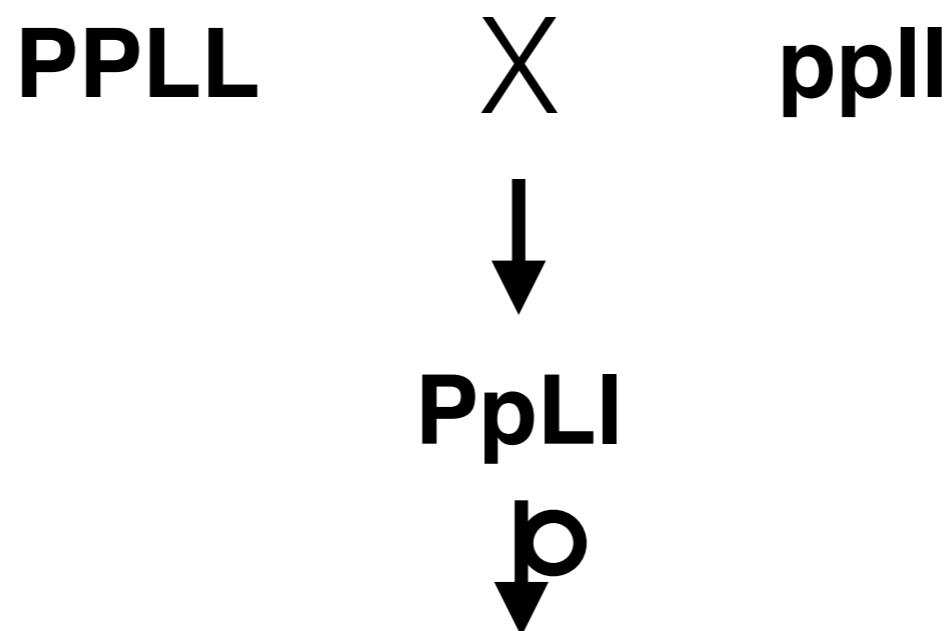
P= purple flower

p= red flower

L= long pollen

l= short pollen

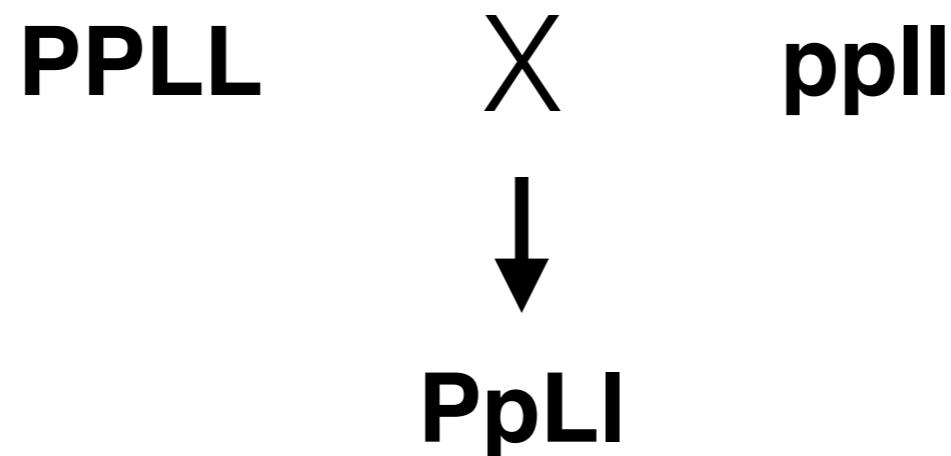
# Bateson and Punnett's pea crosses



Phenotype	Expected number	Expected ratio	Observed number
Purple Long	215	9	284
Purple short	71	3	21
red Long	71	3	21
red short	24	1	55

P= purple flower  
p= red flower  
L= long pollen  
l= short pollen

# Bateson and Punnett's pea crosses



Parental = allelic combination found in parents  
(most abundant classes, always paired)

Recombinant = allelic combination NOT found in parents  
(least abundant classes, always paired)

# Bateson and Punnett's pea crosses

PPLL      X      ppll



PpLl

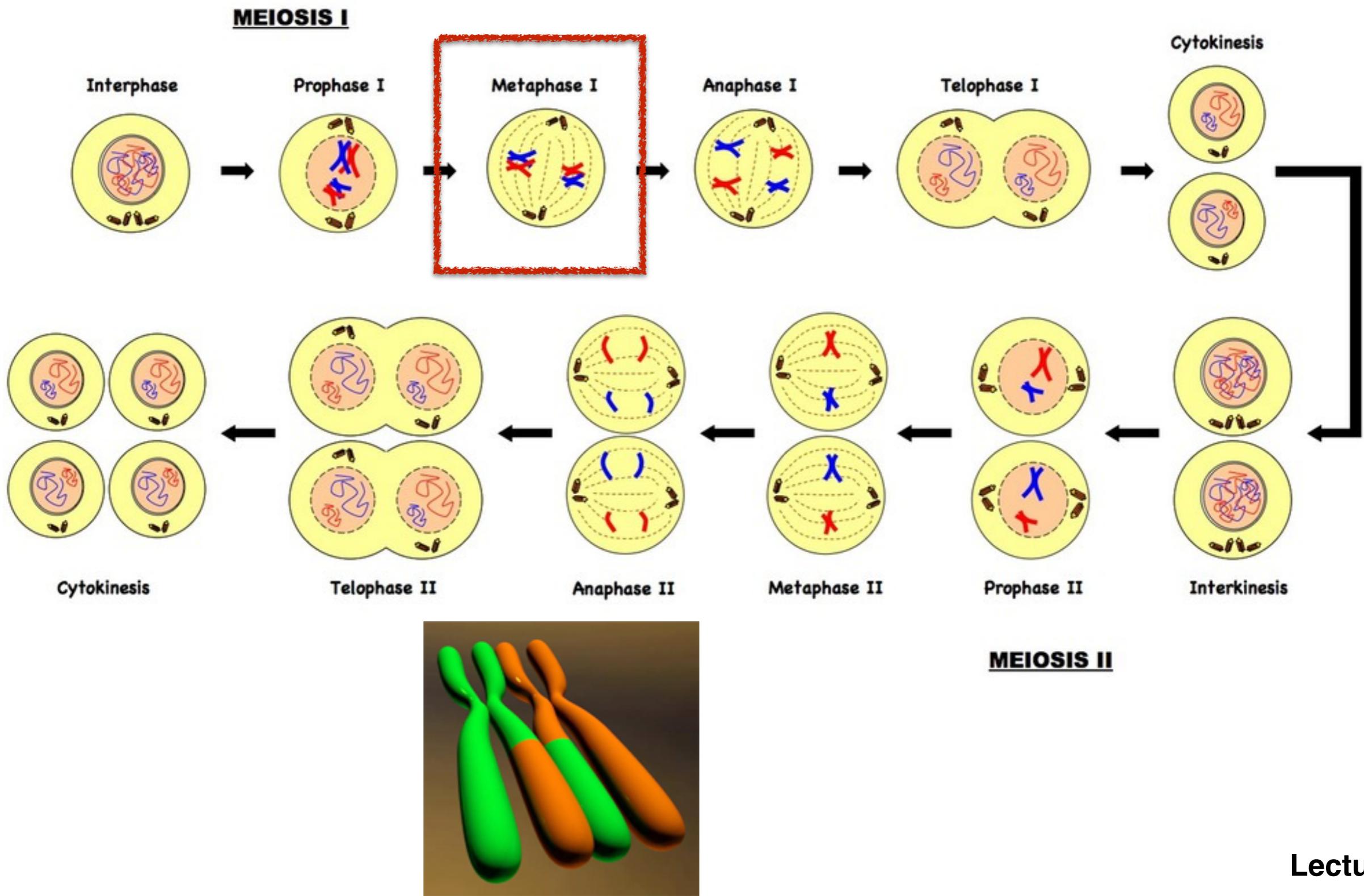


Phenotype	Expected number	Expected ratio	Observed number
Purple Long	215	9	284
Purple short	71	3	21
red Long	71	3	21
red short	24	1	55

P= purple flower  
p= red flower  
L= long pollen  
l= short pollen

Which are recombinant  
and parental offspring?

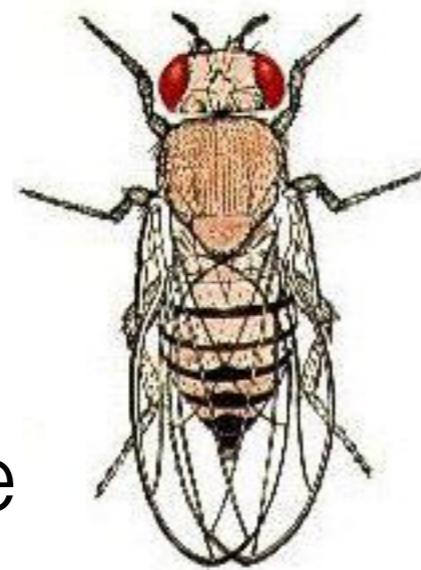
# Meiosis: A reductional division in two acts



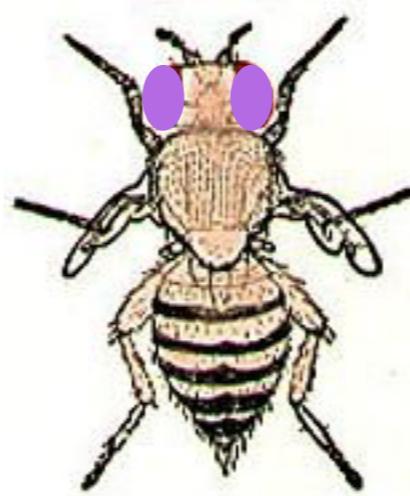
# The fly room at Columbia



Wild-type

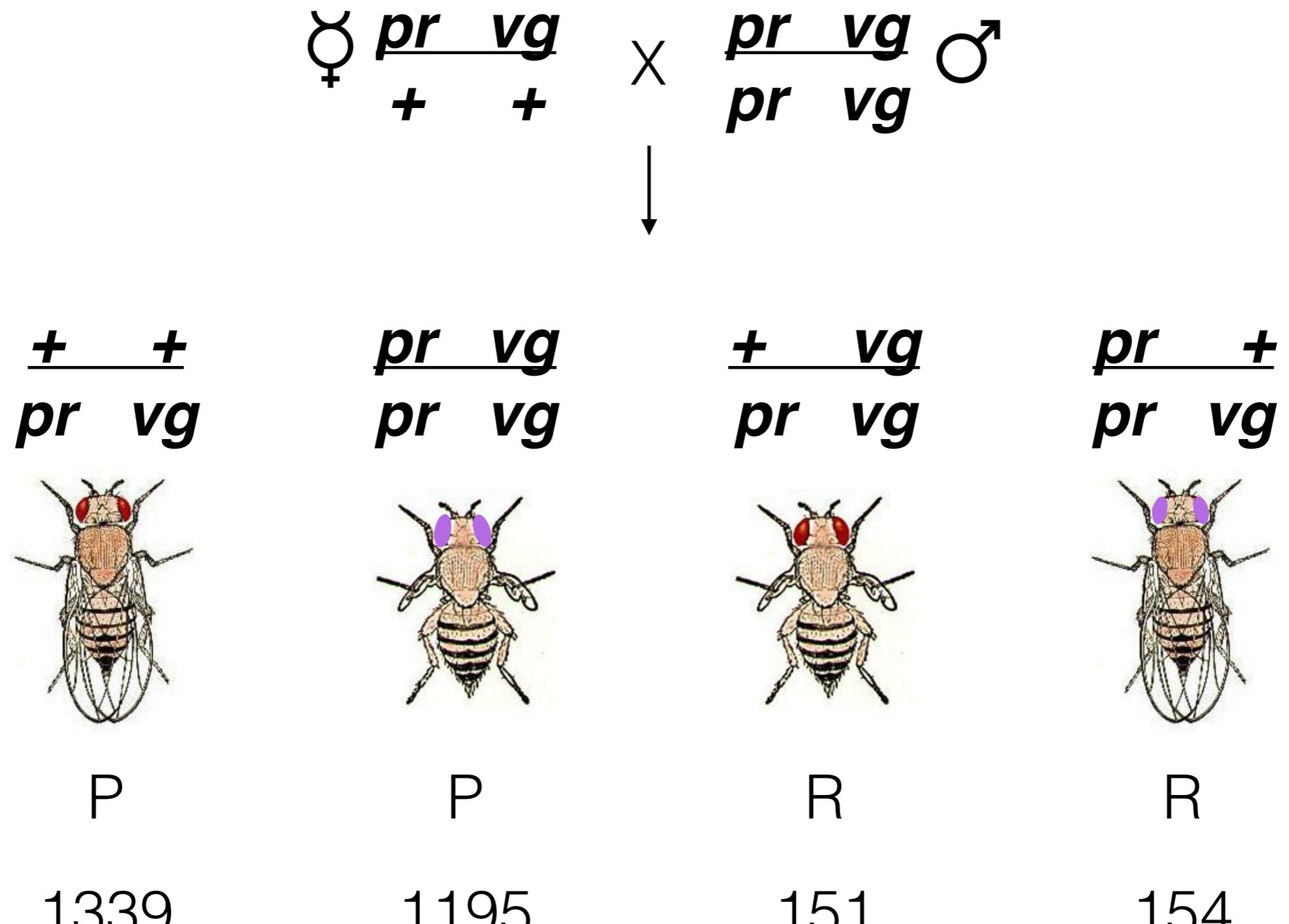


**$+$  +**



Purple eyes,  
vestigial wings

**$pr\ vg$**



**Expectation is equal proportion of each class**

Total = 2839

Lecture 2

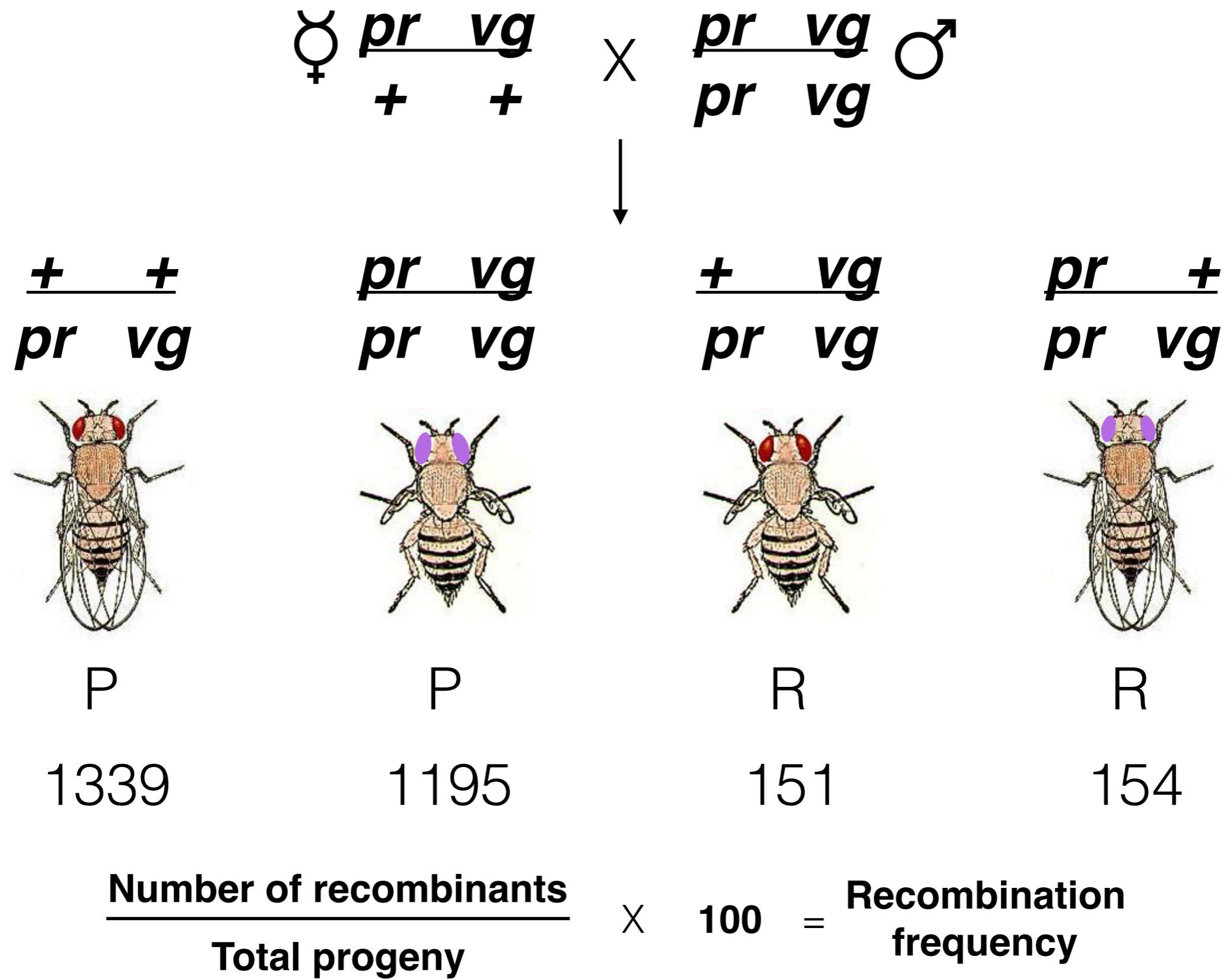


© Copyright California Institute of Technology. All rights reserved.  
Commercial use or modification of this material is prohibited.

**Alfred Sturtevant**

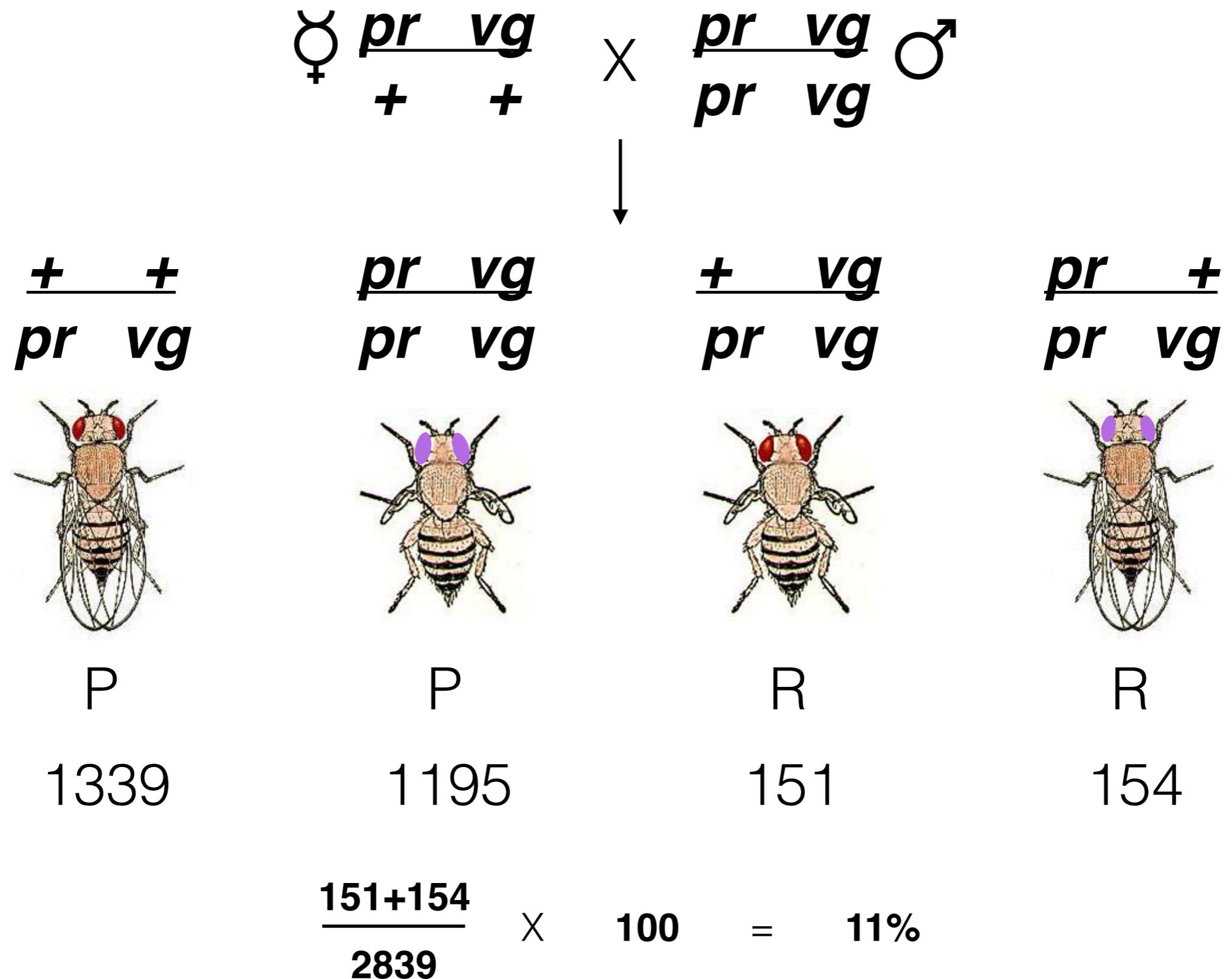
$$\frac{\text{Number of recombinants}}{\text{Total progeny}} \times 100 = \text{Recombination frequency}$$

**1% RF = 1 map unit = 1 centiMorgan**



Total = 2839

Lecture 2



Total = 2839

Lecture 2

# Recombination is the exchange of genetic material between homologous chromosomes

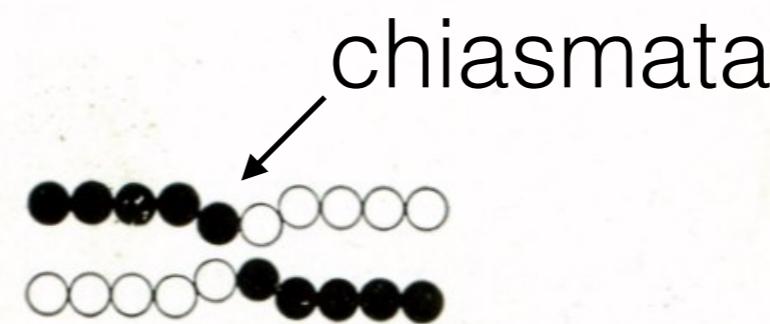
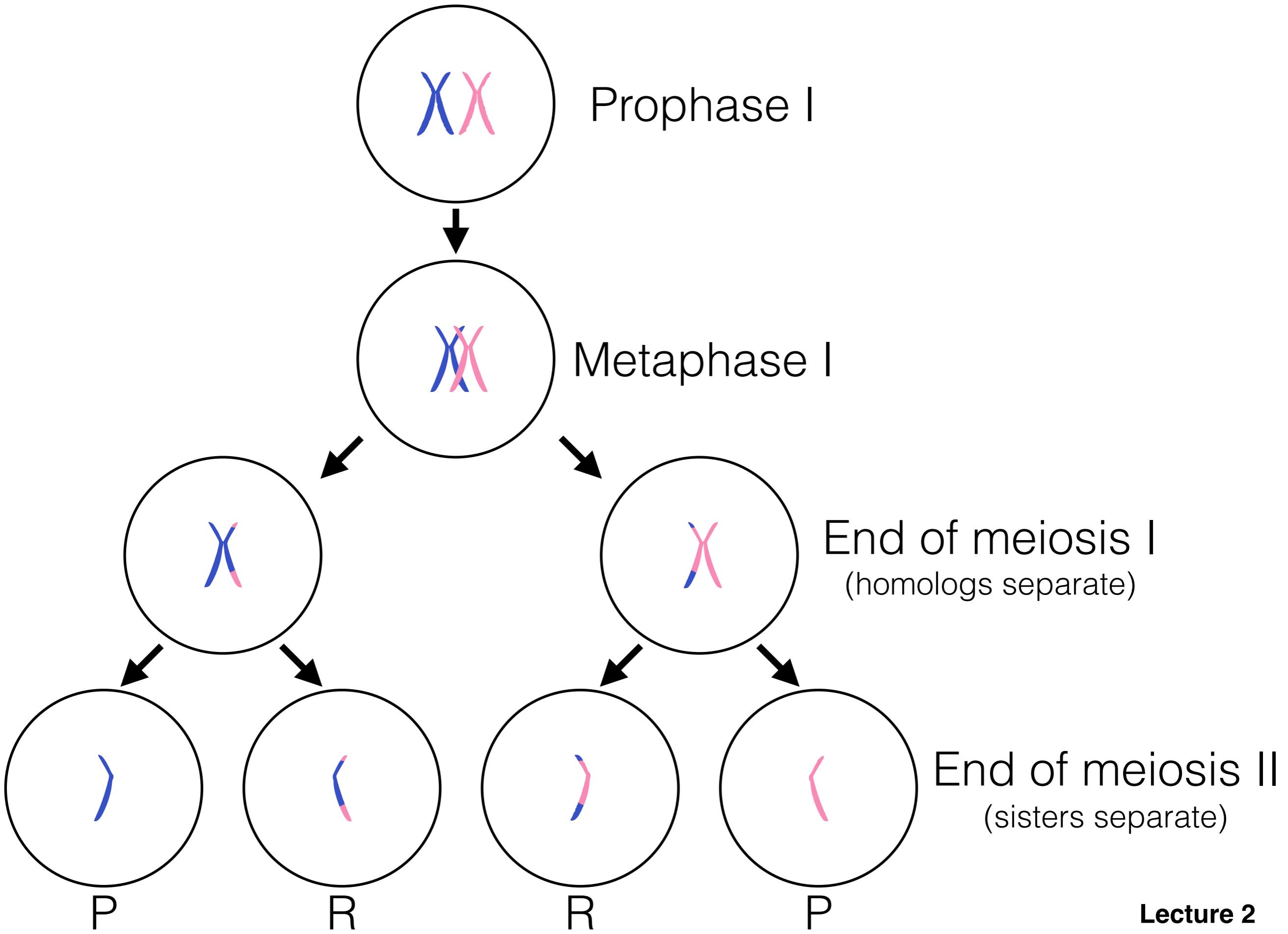
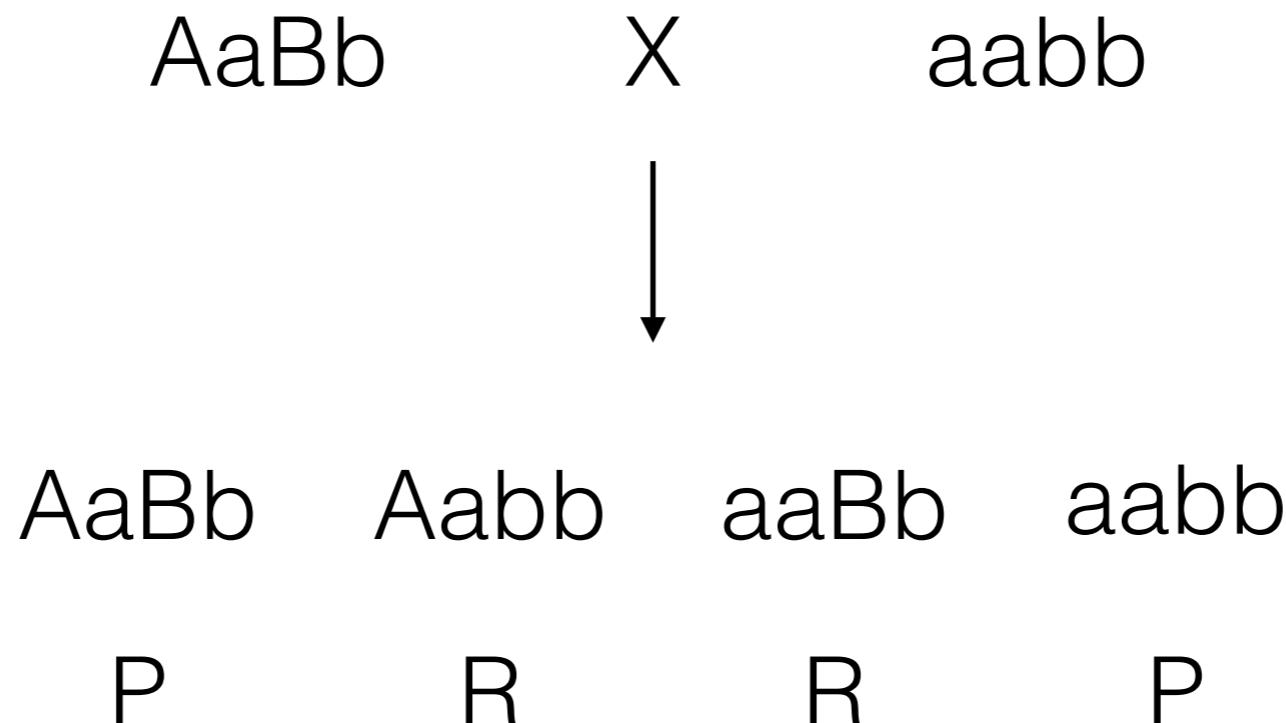


FIG. 64. Scheme to illustrate a method of crossing over of the chromosomes.



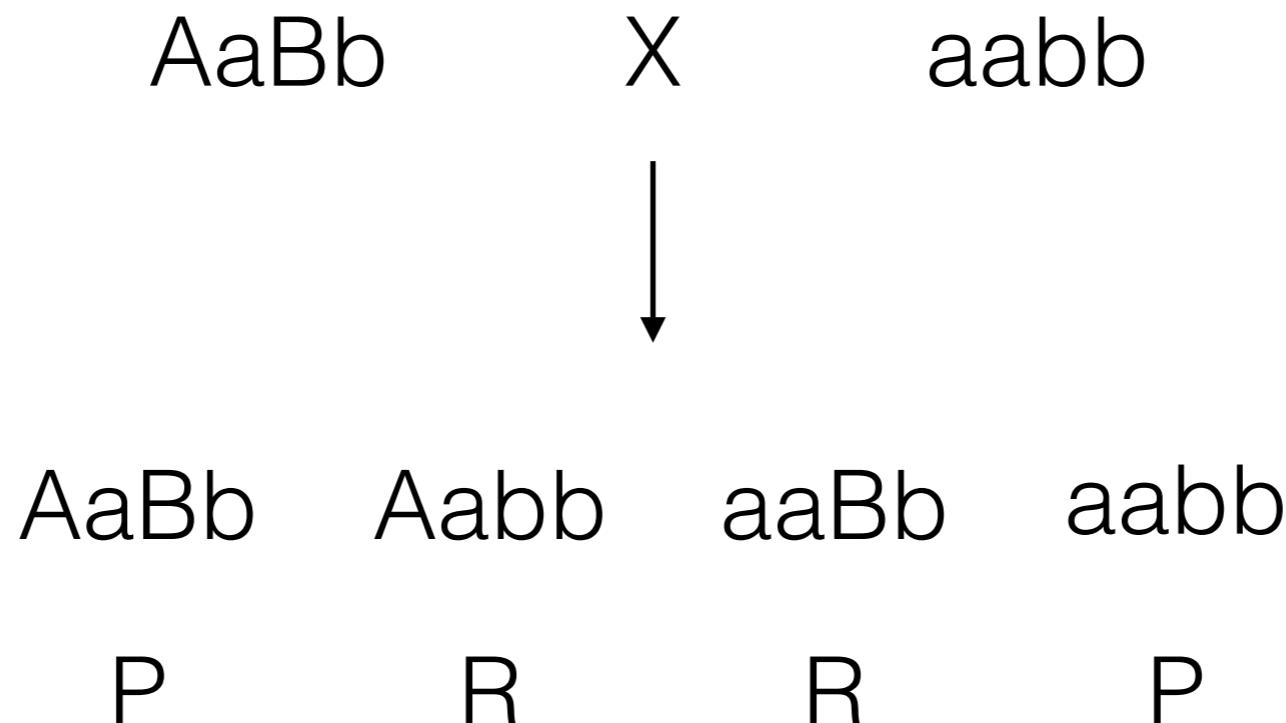
## Independent assortment defines the limit of linkage



All four classes occur in equal ratios

$$\frac{\text{Number of recombinants}}{\text{Total progeny}} \times 100 = \text{Recombination frequency}$$

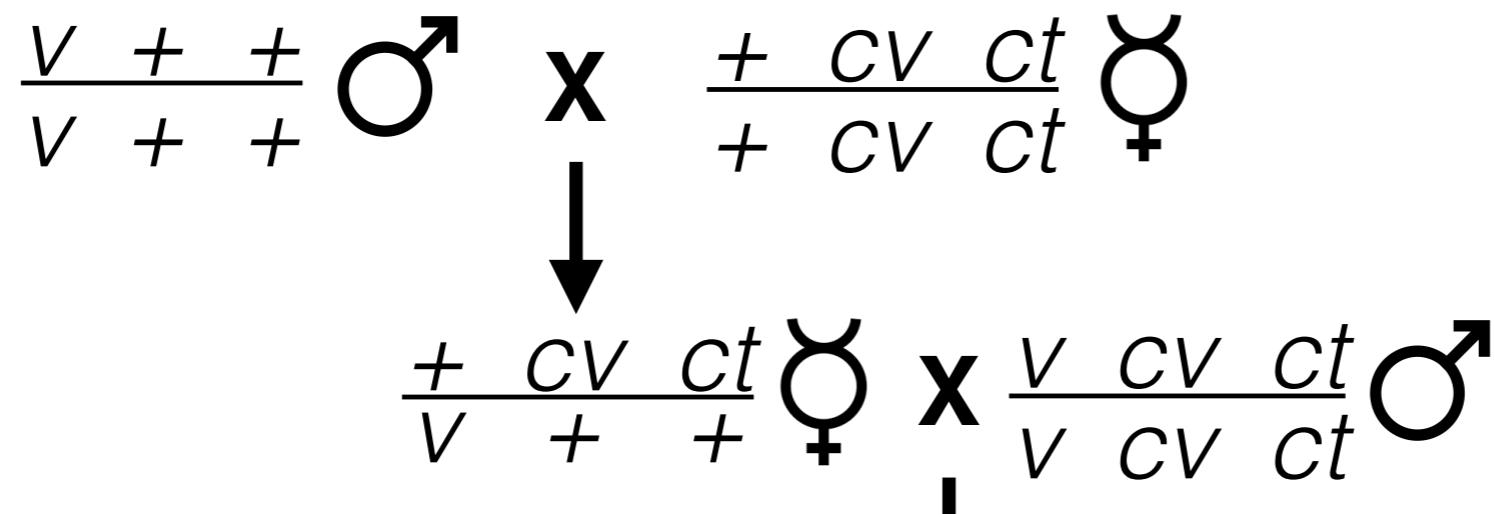
## Independent assortment defines the limit of linkage



All four classes occur in equal ratios

$$\frac{2^*x}{2^*x + 2^*x} \times 100 = 50\%$$

# A three-factor cross



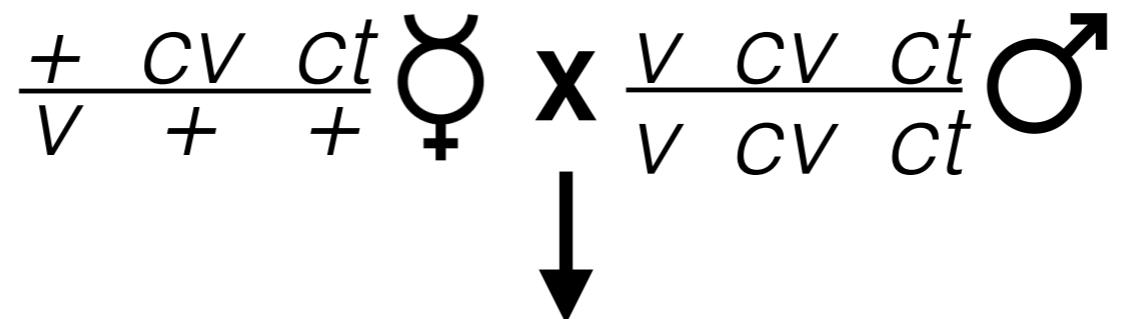
Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

v = vermillion eyes

ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

$+ \quad CV \quad ct$       **P**  
 $V \quad + \quad +$       **P**

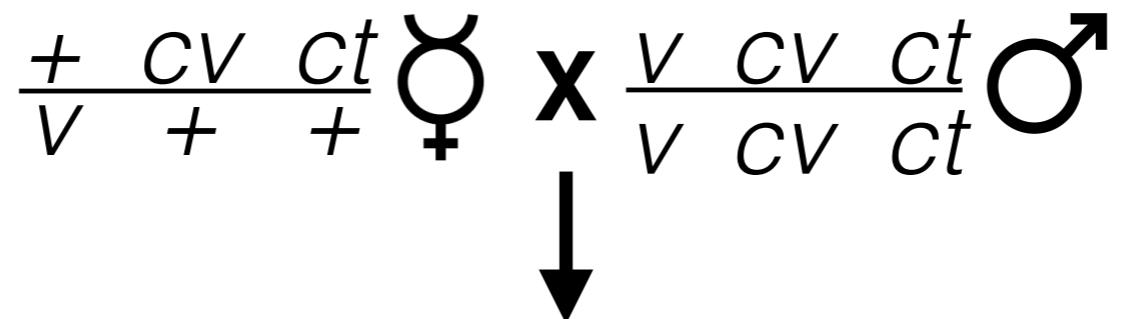
1. Determine parental class, label

$v$  = vermillion eyes

$ct$  = cut wings

$cv$  = crossveinless wings

$+$  = red eyes and normal wings



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

$+ \quad CV \quad ct$       **P**  
 $V \quad + \quad +$       **P**  
**R**  
**R**  
**R**  
**R**  
**R**  
**R**

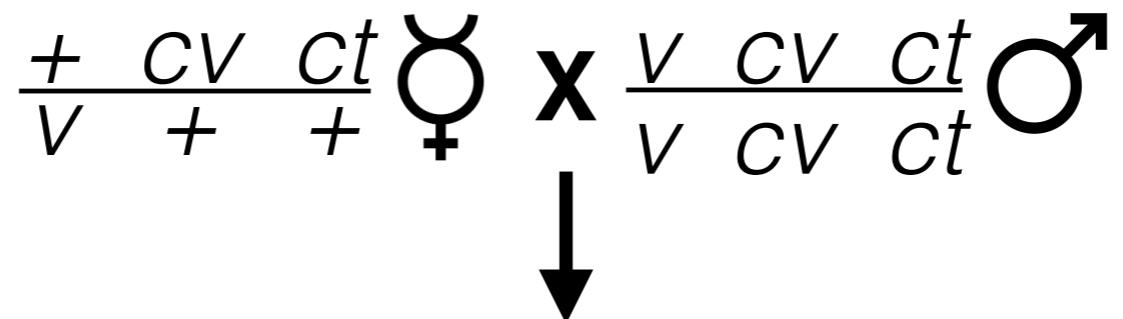
1. Determine parental class, label
2. Are all classes present?

v = vermillion eyes

ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

+ CV ct      P  
 V + +      P  
 R      R  
 R      R  
 R      R  
 + CV +      R  
 V + ct      R

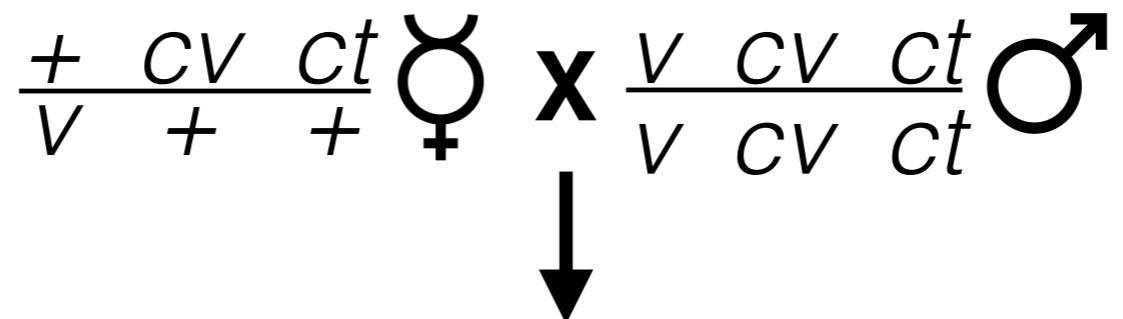
1. Determine parental class, label
2. Are all classes present?
3. Least abundant class is double recombinant, tells gene in middle

v = vermillion eyes

ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

+ CV ct	P
V + +	P
+ + ct	R
V CV +	R
+ + +	R
V CV ct	R
+ CV +	R
V + ct	R

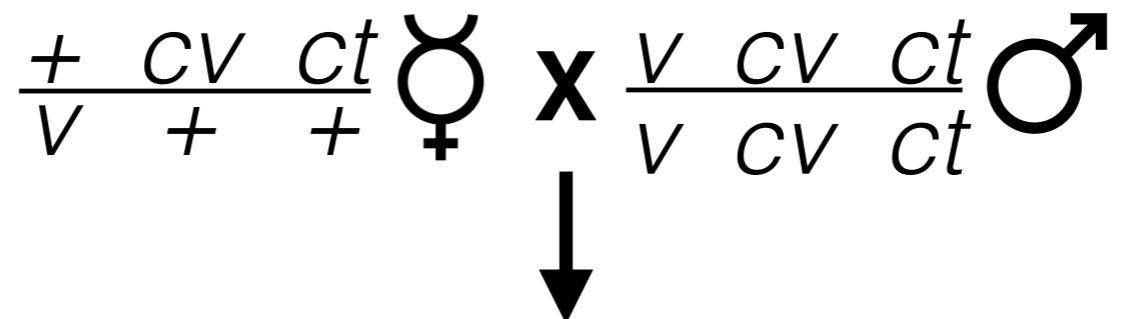
1. Determine parental class, label
2. Are all classes present?
3. Least abundant class is double recombinant, tells gene in middle
4. Write out the genotypes of the offspring

v = vermillion eyes

ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

+ CV ct P  
 V + + P  
 + + ct R  
 V CV + R  
 + + + R  
 V CV ct R  
 + CV + R  
 V + ct R



1448 total progeny

1. Determine parental class, label
2. Are all classes present?
3. Least abundant class is double recombinant, tells gene in middle
4. Write out the genotypes of the offspring
5. Calculate distance from one gene to middle gene **v to ct**

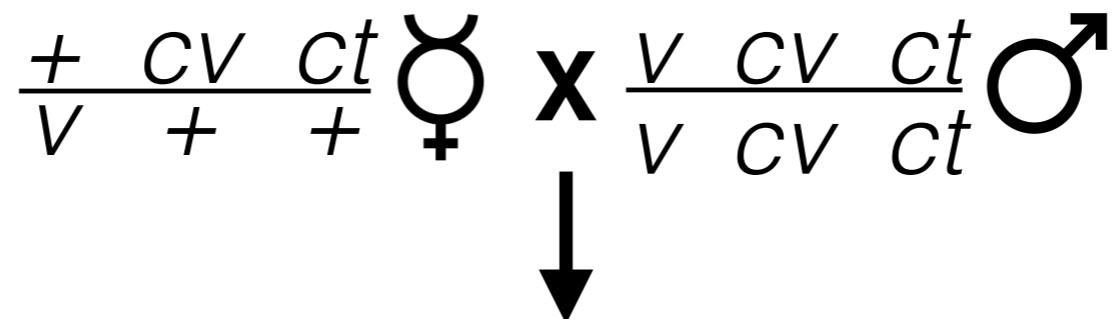
v = vermillion eyes

ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings

$$\frac{94+89+5+3}{1448} \times 100 = 13.2\%$$



Eye Phenotype	Crossvein Phenotype	Cut Phenotype	Number of offspring
Red	No crossvein	Cut wing	580
Vermillion	Crossvein	Normal wing	592
Red	Crossvein	Cut wing	40
Vermillion	No crossvein	Normal wing	45
Red	Crossvein	Normal wing	94
Vermillion	No crossvein	Cut wing	89
Red	No crossvein	Normal wing	5
Vermillion	Crossvein	Cut wing	3

$+ \quad CV \quad ct$  P  
 $V \quad + \quad +$  P  
 $+ \quad + \quad ct$  R ←  
 $V \quad CV \quad +$  R ←  
 $+ \quad + \quad +$  R  
 $V \quad CV \quad ct$  R  
 $+ \quad CV \quad +$  R ←  
 $V \quad + \quad ct$  R ←

1448 total progeny

1. Determine parental class, label
2. Are all classes present?
3. Least abundant class is double recombinant, tells gene in middle
4. Write out the genotypes of the offspring
5. Calculate distance from one gene to middle gene
6. Calculate distance from the other gene to middle gene

**cv to ct**

v = vermillion eyes

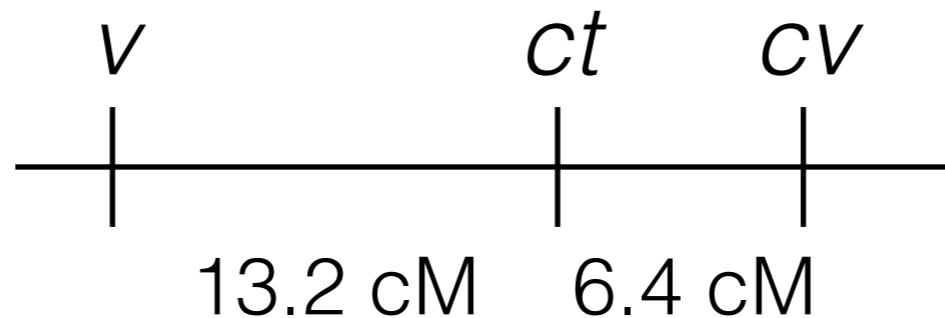
ct = cut wings

cv= crossveinless wings

+ = red eyes and normal wings

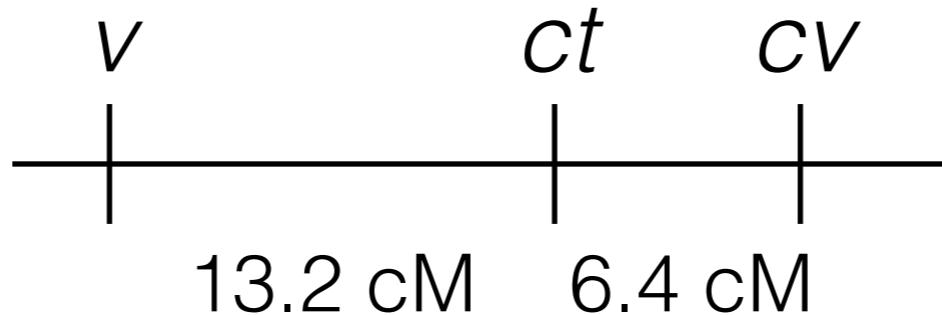
$$\frac{40+45+5+3}{1448} \times 100 = 6.4\%$$

# Our first genetic map



1. Order by least abundant class
2. Arbitrary which genes on ends
3. Class *v* to *cv* undercounts because double recombinants look like parentals

## Our first genetic map

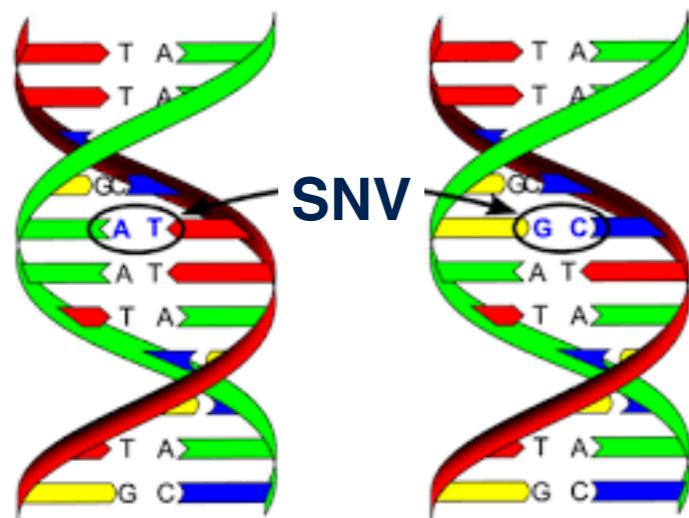


1. Order by least abundant class
2. Arbitrary which genes on ends
3. Class *v* to *cv* undercounts because double recombinants look like parentals

**We have a better way!**

# Molecular markers are often used for genetic mapping

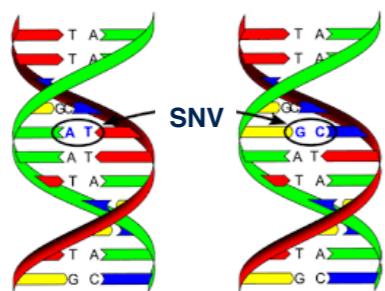
- Single nucleotide variants



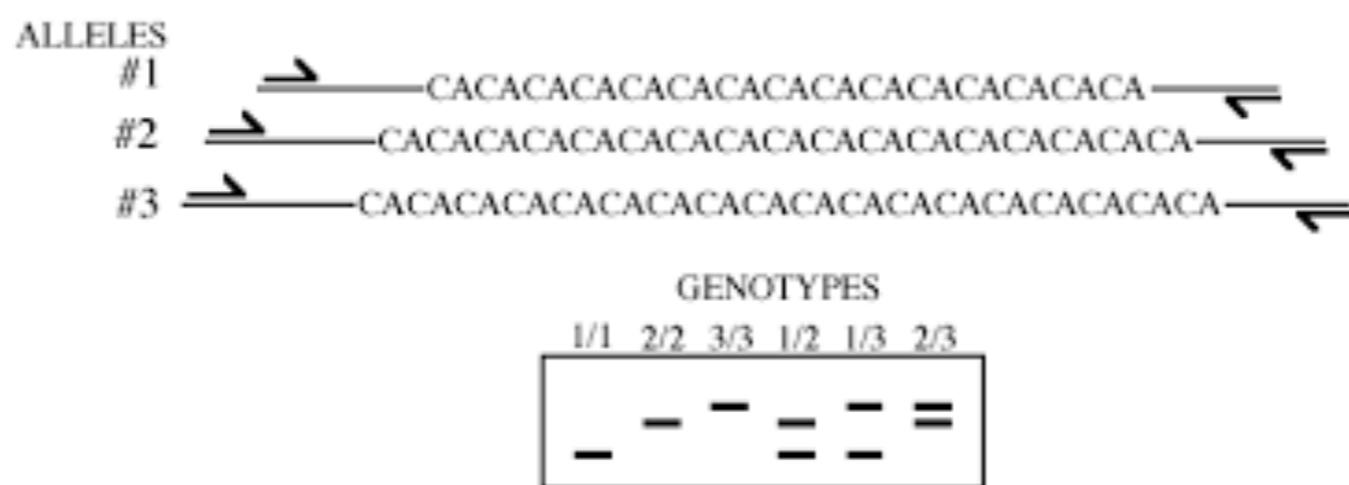
Detected by sequencing,  
hybridization (array), or PCR.

# Molecular markers are often used for genetic mapping

- Single nucleotide variants

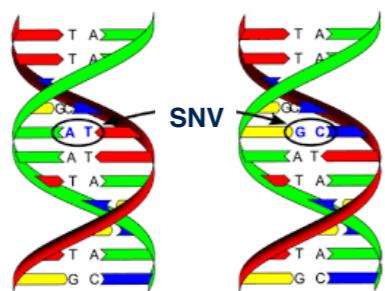


- Microsatellite repeats



# Molecular markers are often used for genetic mapping

- Single nucleotide variants

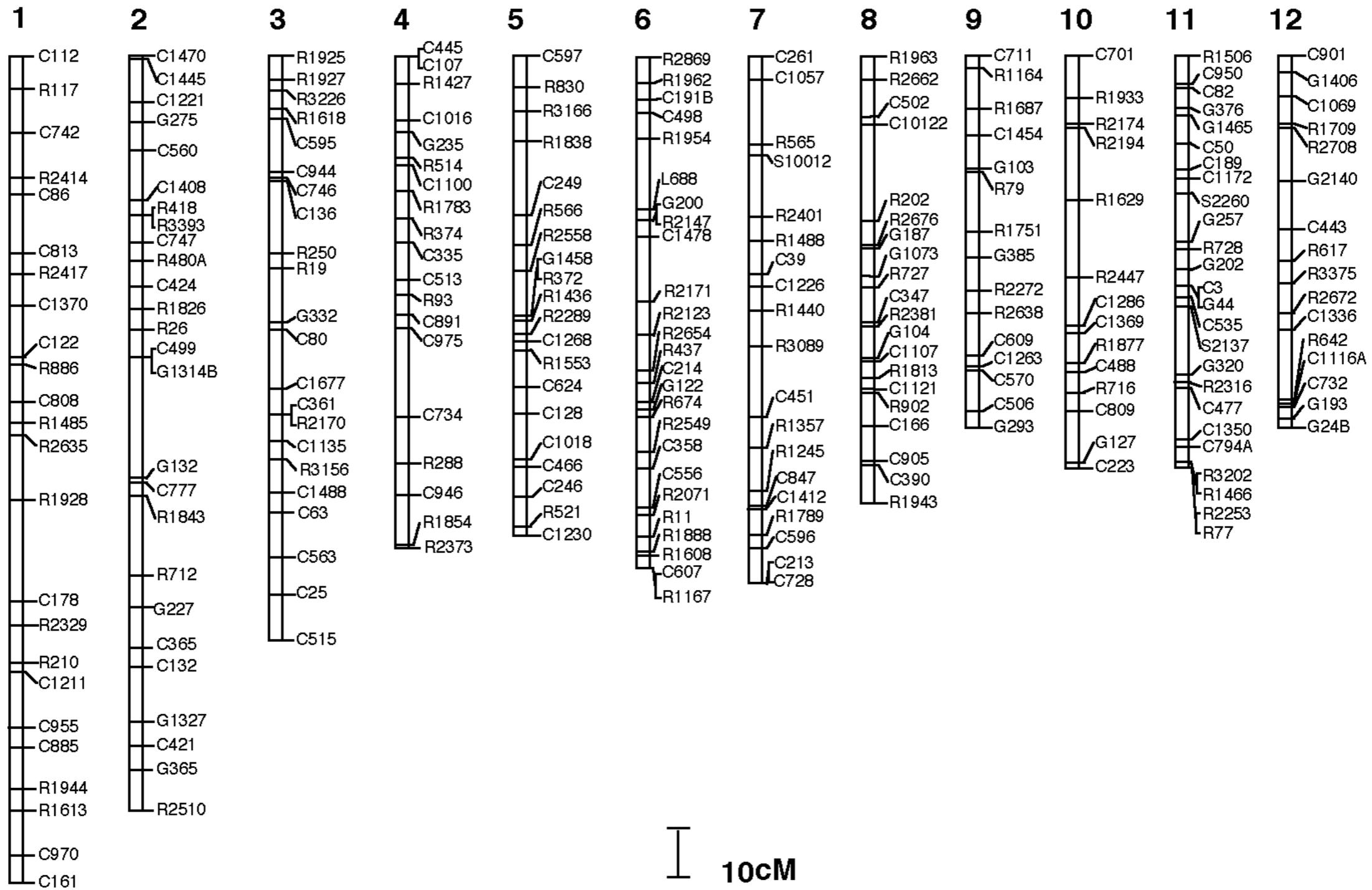


- Microsatellite repeats



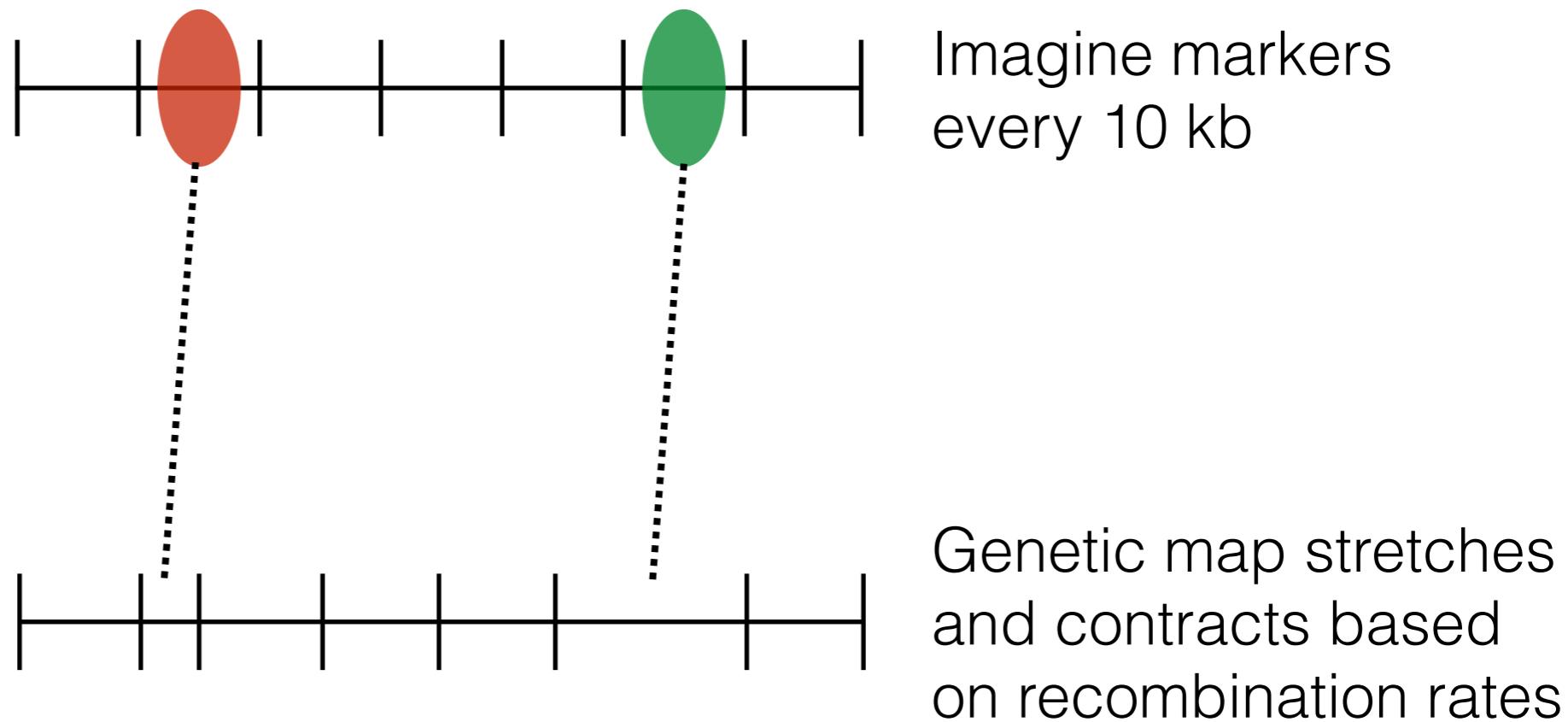
- Insertion/deletion variants

# Molecular markers are used most of the time



10cM

# What do regions of more or less recombination do to the linkage map?

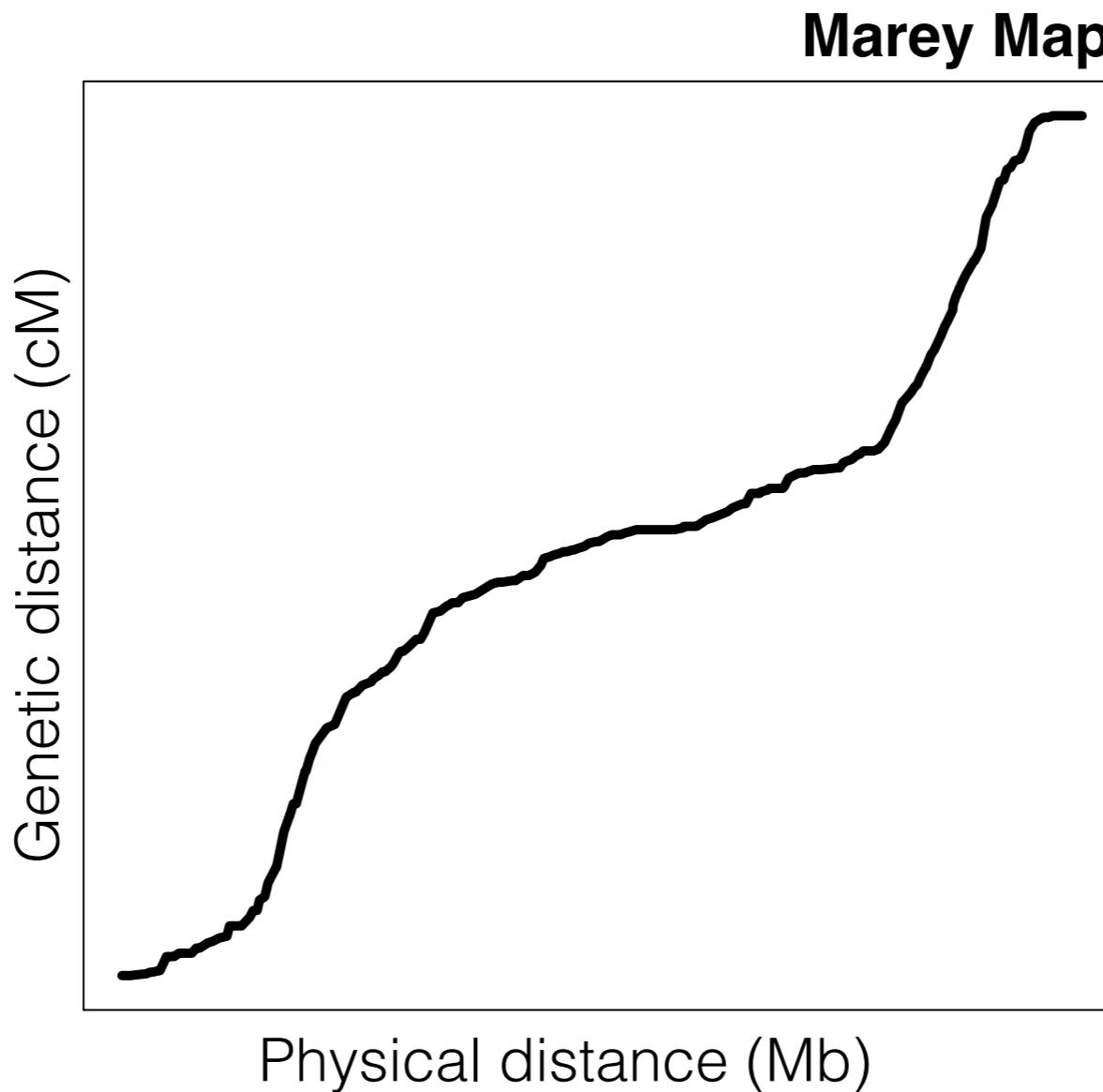


Less recombination (cold spot)



More recombination (hot spot)

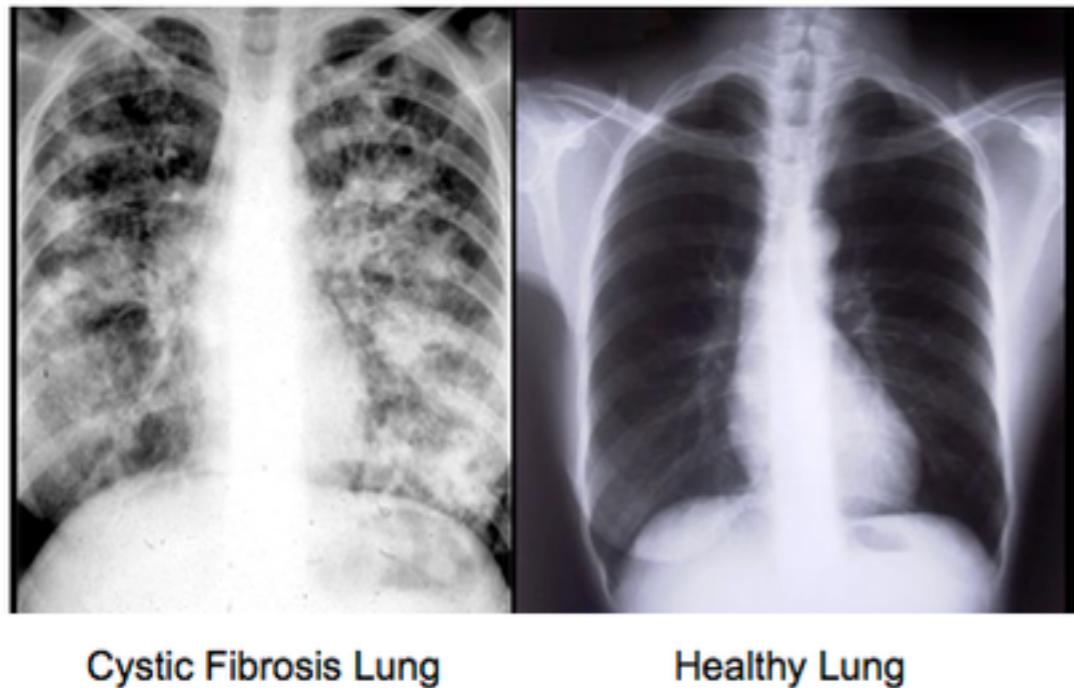
# What do regions of more or less recombination do to the linkage map?



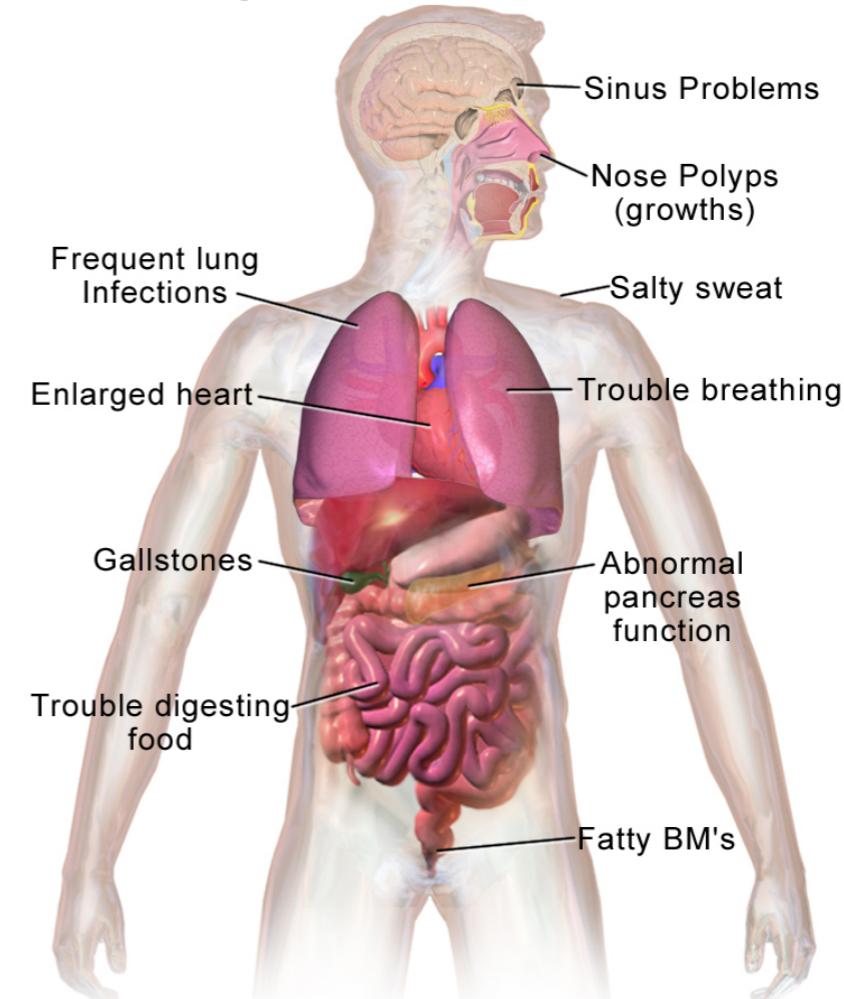
**Why do we care?**

**Is this technique “old-fashioned”?**

# What about cystic fibrosis and today's topic?



## Health Problems with Cystic Fibrosis



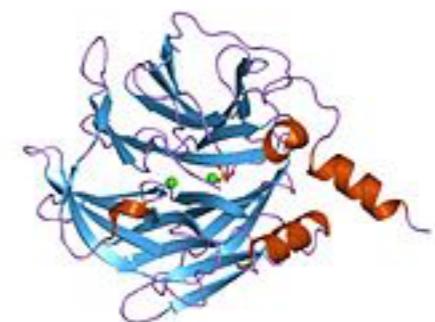
Where is the gene that when mutated causes cystic fibrosis?

How would you map it?

# Linkage to allozymes

Allozymes are enzymes with activities that vary from person to person

*PON1* is a hydrolase used for detoxifying cells.  
Activity varies from person to person.



68 families with at least two children with and without CF were phenotyped for *PON1* activity and CF.

Found linkage to *PON1* and chromosome 7

# What have we learned so far?

CF is rare – 1/10,000 births

Autosomal recessive disorder

Not caused by chromosomal aberrations or NDJ

Linked to *PON1* on chromosome 7

