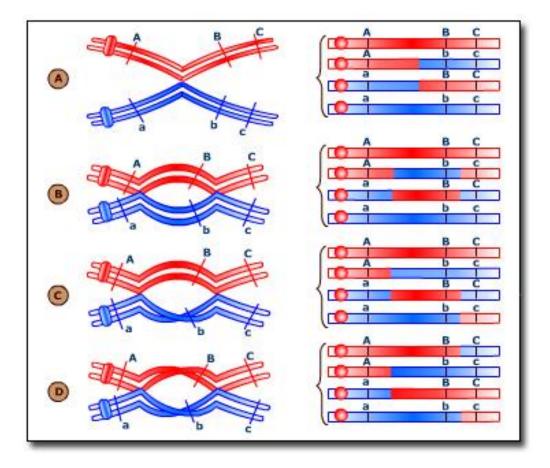
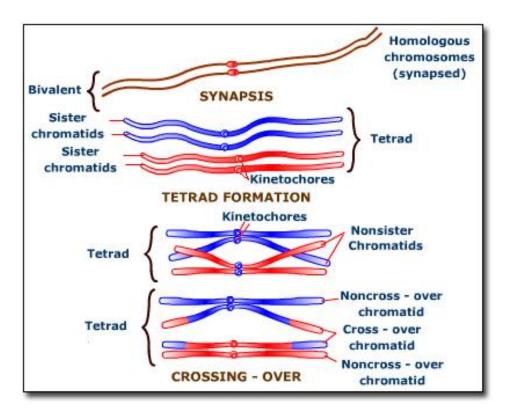
## **CROSSING OVER - FACTORS AFFECTING IT**

## Crossing over

Genetic recombination crossing over is the mutual exchange of the corresponding parts of the adjacent paternal and maternal chromatids of the pachytene of meiosis I, producing new combinations of genes. The chromatids resulting from the interchange of segments are known as the cross over recombinants and the chromatids that remain intact are called non-crossover parental chromatids.



A :Single Crossing, B :Reciprocal Double Crossing Over, C and D: Complementary Double Crossing Over



# It produces 50% parental and 50% recombinant chromatids

# Significance of Crossing Over

a. It produces new individuals having new combinations of traits.

b. Crossing over has helped in establishing the concept of linear arrangement of genes.

c. The frequency of crossing over helps in the mapping of chromosomes. i.e., determining the location of the genes in the chromosomes.

d. Selection of useful recombination by geneticists has brought about green revolution in our country.

## Stern's Drosophila Experiments

In 1931, a few weeks after publication of similar experiments by Creighton and McClintock in *Zea mays* Stern reported work with *Drosophila*.

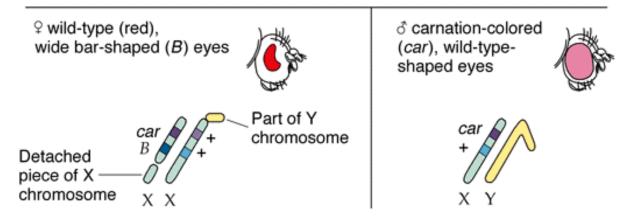
<sup>9</sup> wild-type (red), wide bar-shaped (B) eyes



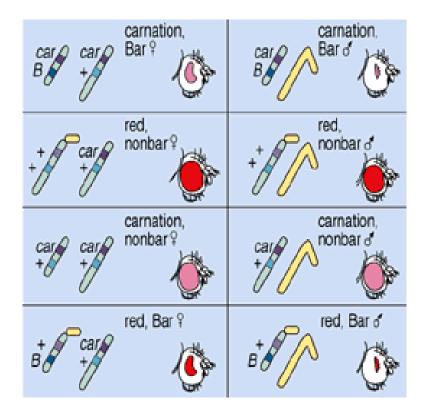
ੈ carnation-colored (*car*), wild-typeshaped eyes

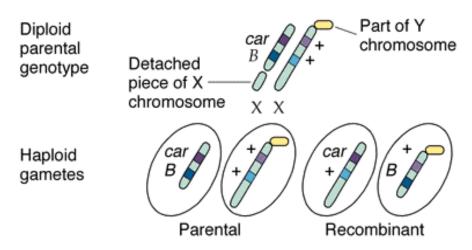


There are 2 Linked traits: carnation eye and bar-eye flies.Carnation is recessive, bar-eye is dominant. In B/B homozygotes, the eye is very narrow. In B/+ heteros, get "wide-bar".



Male had normal X chromosome, car +/Y and Female had 2 cytologically distinct X chromosomes. One X had a portion of the Y chromosome attached to it, making it longer than normal. This one was wild type for both genes. The other X was shorter than normal, and had the *car* and *B* alleles on it.





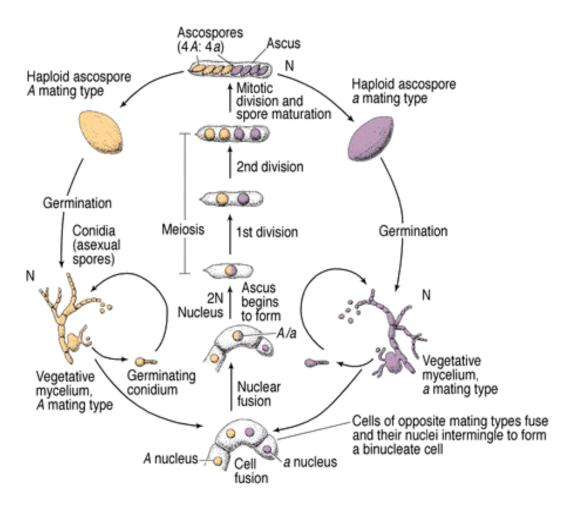
The observed results require the above types of gametes to be produced by females. The key observation was that in every case of genetic recombination, there had also been recombination of the cytological features. The same type of results had been found in the corn experiments. Genetic recombination (phenotypes of the offspring) was always accompanied by recombination of the cytological features. Together, these two studies convinced the field of genetics that

physical exchange of chromosome segments was the basis for recombination of linked genes.

BUT, when did the crossing-over take place. Remember, at the start of meiosis, the homologues are already replicated, and already paired up. So, one plausible model is that crossing-over actually takes place during interphase, before replication. That is, when each chromosome pair would consist of 2 chromatids, not 4 (not the tetrad stage).

How to distinguish between the two models?

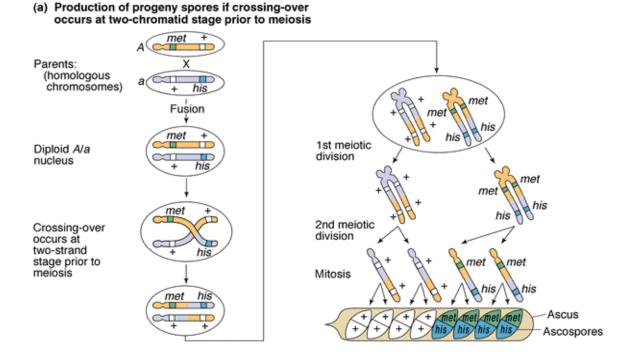
To distinguish between the two models a right model organism Neurospora crassa (a bread mold) was selected.



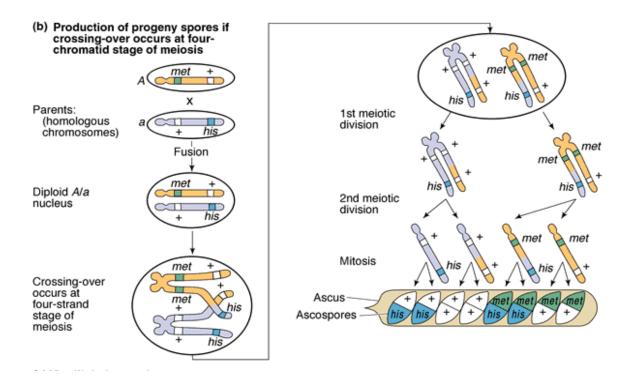
Normally, it reproduces asexually and also sexually. There are two mating types: *A* and *a*. The 2 haploid nuclei fuse to give a diploid nucleus, the only diploid

phase to this life cycle. Meiosis produces 4 haploid spores within an ascus. Subsequent mitotic division produces 8 haploid, sexual ascospores in a linear arrangement. All the products of a single meiosis are found together in the ascus. and, the order of the 4 spore pairs is the same as the order of the 4 chromatids of each tetrad during Metaphase I. By capitalizing on the biology of our model organism, we can now rigorously test our 2 competing hypotheses: Does crossing over take place during the 2-chromatid phase, or during the 4-chromatid phase?

To test, we need strains with mutations at 2 genes on the same chromosome. In *Neurospora*, there are many <u>nutritional mutants</u> are known. These are unable to make some essential nutrient on their own, and it must be supplied in their medium. *met* mutants are unable to synthesize their own methionine. *his* mutants are unable to synthesize their own histidine.



All spores are recombinants.



Note: 1/2 are parental, 1/2 are recombinant. Repeated experiments like this have shown that we get the second type of result. I.e., crossing over takes place at the tetrad stage (4 chromatids). They also show that the exchange is <u>reciprocal</u>: equal amounts of the 2 chromosomes are swapped.

## CHROMOSOME MAP

## (Linkage map / Cross over map / Genetic map)

The chromosome map may be defined as a line, on which the genes are represented by points, separated by distances proportional to the amount of crossing over.

The chromosome maps are also referred to as cross over maps since they are sketched by the amount of crossing over.

The percentage of crossing over is directly proportional to the distance of the alleles showing crossing over in the chromosome.

The chromosomes maps are the graphic representation of the genes in a chromosome.

The percentage of crossing over is calculated by test crosses. In mapping the genes, a unit of distance is used and it is called as *map unit* or *Morgan unit*.

The first chromosome map was made in 1911 by *Sturtevant* and soon after additional maps were made by *Bridges* and others.

Drosophila is the earliest material used by the scientists, for constructing maps.

#### Procedure for the chromosome mapping

In fact genes are plotted on the chromosome on the basis of crossing over results between different pairs of linked genes. The actual distance between two genes is said to be equivalent to the percentage of crossing over between these genes.

When the % of crossing over between two genes is 5, then the distance is 5 units. For example five genes A, B, C, D and E are to be plotted on a chromosome. If cross over results indicate that genes A and E have the highest percentage of crossing over, it means that these should be placed at the maximum distance.

In this example, the gene A can be taken as a starting point in the chromosome and can be represented by O.

Now if the gene A and B exhibit 7% crossing over, the gene B can be placed on the chromosome at a distance of 7 units.

If the gene C shows 8% crossing over with gene B and about 15% crossing over with gene A, it can be plotted on the chromosome at a distance of 15 units from gene A.

Similarly if gene A and E exhibit 20% and 30% crossing over with gene D and 5% and 10% with gene C these, are located on the chromosome 5 and 10 units away from the gene C respectively.

#### Construction of Chromosome map in Drosophila

In Drosophila the chromosome map is constructed with the help of test cross. In Drosophila grey colour is dominant over black colour; and the long wing is dominant over vestigial wing.

The  $F_1$  female hybrid is **test crossed**. Four types of individuals are formed. Out of four types, two types are parental type (G:L & B:V) and other two are non parental type (G:V & B:L) due to crossing over. Non – parental type is 17%. So the percentage of crossing over is equivalents to 17%. The distance between the two genes (G-L) is equivalent to the percentage of crossing over or percentage of non parental combination. So the distance between the gene G & L is equivalent to 17 morgan units.

Percentage of non parental combination	=	17%
So the percentage of crossing over	=	17
So the distance between the Gene G & L	=	17 map unit

In another experiment the  $F_1$  female grey red is test crossed with black cinnabar. The experiment shows 9% non parental combination individuals. So the distance between the Gene G & Cn is equavalent to 9 map unit.

In the same way the  $F_1$  female red long is test crossed with cinnabar vestigial. The experiment shows 9.5% non-parental combination individuals. So the distance between the gene Cn is equivalent to 9.5 map unit.

According to the first experiment the distance between G & L is equivalent to 17 map unit. But the second and third experiment show 18.5 map units between the two genes. To find out the actual reason for this difference in the distance, conduct a 3 point cross.

#### **Three Point cross**

In the three point cross all the three pairs of genes are considered in the experiment. The  $F_1$  hybrid female is test crossed. They produce 8 different types of individuals. Out of 8 types, two types are parental. Remaining six are non-parental.

	Male		Female
Parent :	Grey Red Long		Black cinnabar Vestigial
	G Cn L	х	<u>g cn l</u>
	Y-chromosome		g cn l
		Norma	al
F <sub>1</sub> :		<u>G Cn I</u>	L
		g cn l	
Back cross :	Female		Male
	Normal		Recessive
	<u>GCnL</u> x		<u>g cn l</u>
	g cn l		Y – chromosome

# F<sub>2</sub> Offspring

Parental combinations due to linkage = 658 = 82.25%

Non parental combination : Single cross over :

a) Between G and Cn 1. Black red long g cn l 2. Grey cinnabar vestigial G cn l = 34g cn l

b) Between Cn and L

1. Grey red vestigial <u>G Cn I</u>

Double cross over

1. Grey cinnabar long	<u>G cn L</u>	=_ 4	
g cn l		= 6	
2. Black red vestigial <u>g Cn I</u> = 2			

q	cn	I
0		

Total c.o between G and Cn	=	70 + 6 = 76 = 9.5%
(includes double c.o)		
Total c.o between Cn and L	=	66 + 6 = 72 = 9.0%
(includes double c.o)		
Total c.o. between G and L	=	76 + 72 = 148 = 18.5%
(includes double c.o)		
C.o between G and E	=	70 + 66 = 136 = 17%
(double c.o not included)		

From these results, it is concluded that the gene cinnabar lies about half – way between the genes for black body colour and vestigial wings. The total amount of crossing over between black body and vestigial wing is 18.5% rather than the 17% expected on the basis of the first cross. The discrepancy (18.5 - 17 = 1.5) just noted, arises because of the occurrence of double crossing over, that is, of two cross overs occurring simultaneously in the same cell between these two loci.

As a final check on these results, it would be well to make a trihybrid or three point cross using all three pairs of genes at once. When pure recessive flies are crossed with normal flies, all the  $F_1$  flies are normal phenotypically. When the  $F_1$  females are back crossed to triple recessive males, eight phenotypes are obtained.

From the data obtained the relative position of the genes can be calculated. The distance between G and Cn is 9 units, the distance between Cn and L is 9.5 units; the L gene could be to the right of Gn locus or to the left. If the first order (CnL) is correct, then the distance between G and L is 17 units. This small discrepancy is due to double crossing over. Based upon the above data, the three genes can be mapped as follows :

#### **Chromosome Maps of Drosophila**

The chromosome maps of Drosophila include four linkage groups corresponding to four chromosome pairs. The genes present in the X chromosome constitute the first linkage group, those present in  $2^{nd}$  and  $3^{rd}$  chromosome constitute  $2^{nd}$  and  $3^{rd}$  linkage groups and those on the fourth chromosome form fourth linkage group. The fourth linkage group is the smallest of all.

#### Chromosome maps of maize

Chromosome maps of maize have been drawn by R.A. Emerson. As there are 10 pairs of chromosome 10 chromosome maps are seen.

## Factors affecting the mapping

Chromosome map can be constructed only with the help of crossing over percentage. The crossing over percentage is highly modified by the interference and coincidence.

#### Interference

Normally the double crossing over frequency is very low. Because the crossing over and chiasma formation in the homologous non sister chromatids interferes with the crossing over and chiasma formation at other points nearby. This is called as *interference*. This was discovered by Muller (1911). The interference is *inversely proportional* to the crossing over percentage. The

interference is maximum over a short distance and decreases as the distance increases.

## Coincidence

The coincidence is an inverse measure of interference. It is measured as a ratio between actual number of double cross overs and the expected number of double cross overs.

## Actual number of double cross overs

Coindicidence = Expected number of double cross overs

If the actual number of double cross overs is zero, then coincidence is zero and interference is complete. If the actual number of double cross overs is the same as the expected number, coincidence is said to be one, and interference is nil. It ranges from 0 to 1.