

Chapter 1→Genetics and the Organism (8-31-05)

Genetics→Study of gene function, from molecules of DNA to the gene pool of a population.

DNA contains ALL of the information for building an organism.

Central Dogma of Molecular Biology (Information Flow)

DNA→RNA→Protein

Genes→Functional units of DNA.

Individuals inherit genes.

Individuals contribute to the gene pool of a population.

Alleles→Different, but related, forms of the same gene.

Genotype→The set of genes an organism inherits.

(Very little change over time)

Phenotype→Characteristics of an organism.

Phenotypes change throughout the life of an organism as its genes interact with the environment.

Phenotype is determined by the genotype as it interacts with the environment.

Interactions between genes and the environment determine what organisms become.

Geneticists study partial genotypes and partial phenotypes of organisms.

(i.e. study the effect of one or a few genes)

A particular phenotype can be caused by more than one genotype.

A/A→dominant phenotype

A/a→dominant phenotype

a/a→recessive phenotype

Wild Type (WT)→genotypic or phenotypic characteristics of natural populations or standard laboratory strains.

An individual that is not WT is a variant or mutant.

Mutation is the basis for variation within a species and is the raw material for evolution.

Model Organisms

Genetic analysis is greatly simplified by using an easily followed phenotype in an organism that is practical to study. (e.g. mice vs. elephants)

1. Bacteriophage and viruses
2. Bacteria (*E. coli* and *B. subtilis*)
3. Yeast (Bread and Beer!!)
4. *Caenorhabditis elegans* (round worm)
5. *Drosophila melanogaster* (fruit fly)
6. Maize (corn)
7. *Arabidopsis thaliana* (small plant)
8. *Mus musculus* (mice)

Developmental Noise → Random molecular events in cells of a developing organism that cause phenotypic variations.

Thus, the genotype determines the phenotype; however, expression of the information in the genotype is influenced by the environment, developmental noise, and other genes.

For Genetic Analysis

1. A phenotype that is easy to identify and monitor.
2. A phenotype that is not highly influenced by the environment (control the environment).
3. Try to determine the influence of a single gene.

Genetic Dissection

- Identify a process to study
- Isolate mutants that interfere with the process
- Identify (clone) the WT genes
- Determine the function of the gene products

Chapter 2→Patterns of Inheritance (9-2-05)

Mendelian Analysis→Analyzing hereditary information using Mendel's techniques and principles.

Gregor Mendel→(1860s) Conducted quantitative and systematic studies of inheritance. Proposed the concept of the gene.

Particulate Inheritance→Characteristics determined by discrete units that are inherited intact.

Mendel's Peas→An individual pea plant produces both pollen and eggs. Peas can self-pollinate or be cross-pollinated.

Each pea (individual) results from a separate fertilization event.

Self-pollination (self)→Pollen and egg from the same flower. Both parents have the same genotype.

Cross-pollination (cross)→Pollen from one plant fertilizes an egg from another plant. Parents have different genotypes.

Pure Line→All offspring produced by selfing or crossing individuals within the same line have the same phenotype.

P→Parental generation. (Individuals in the first cross of a particular series of experiments)

F₁→First Filial Generation. (Progeny from the first cross)

F₂→Second Filial Generation. (Progeny from selfing individuals from the F₁ generation)

Dominant→The phenotype that is expressed in the F₁ progeny when two pure lines are crossed.

Recessive→The phenotype that is covered up by the presence of the dominant phenotype. (Reappears in F₂)

Mendel used reciprocal crosses (Figures 2-5 and 2-6).

Experiment 1

P	pure breeding purple flowers	X	pure breeding (Cross) white flowers
		↓	
F ₁	All purple flowers		

Reciprocal crosses gave the same result.
Purple is dominant and white is recessive.

	F ₁	X	F ₁	(Self)
		↓		
F ₂	3:1 ratio of purple:white			

Recessive phenotype remained as a separate heritable trait.
Mendel studied 7 distinct characteristics or phenotypes (Figure 2-4, Table 2-1).

Experiment 2

P	yellow X green	(cross)
		↓
F ₁	All yellow	(self)
		↓
F ₂	3:1 yellow:green	(self)

- 1/3 yellow F₂ always gave rise to yellow.
- 2/3 yellow F₂ gave 3:1 yellow:green ratio.
- 1/1 green F₂ always gave rise to green.

Underlying the 3:1 F₂ phenotypic ratio is a 1:2:1 genotypic ratio of YY:Yy:yy

Mendel proposed that each gene is present twice (gene pair) (Figure 2-7).

Chapter 2→Patterns of Inheritance (9-7-05)

Monohybrid Cross→Analyzing one characteristic (gene).

Dihybrid Cross→Analyzing two genes simultaneously.

Trihybrid Cross, etc...

Dihybrid Cross→Usually has 4 possible phenotypes.

- 1) Both dominant
- 2) One dominant, other recessive
- 3) Other recessive, one dominant
- 4) Both recessive

Phenotypic ratio of F₂ progeny is 9:3:3:1 (Figure 2-10)

Independent Assortment→Different genes assort independently from one another.

Mendel's Second Law→During gamete formation the segregation of alleles of one gene is independent of the segregation of alleles of another gene.

(i.e., gene pairs on different chromosomes assort independently at meiosis)

Probability→The number of times an event is expected to happen divided by the number of opportunities for an event to happen. (i.e.) $p = x/y$

Dice→ $p(\text{of a 3}) = 1/6$

Gametes→if heterozygous Aa: $p(A) = 1/2$, $p(a) = 1/2$

Product Rule→The probability that two independent events will occur simultaneously is the product of their probabilities.

$p(\text{of two 4s}) = (1/6)(1/6) = 1/36$

RrYy: $p(\text{gamete with 2 recessive alleles}) = (1/2)(1/2) = 1/4$

Sum Rule→The probability of either one of two independent events is the sum of the probabilities.

$p(\text{of two 4s or two 5s}) = 1/36 + 1/36 = 1/18$

RrYy: $p(\text{gamete with 2 recessive or 2 dominant alleles}) = (1/2)(1/2) + (1/2)(1/2) = 1/4 + 1/4 = 1/2$

Probability or chance governs the transmission of genes.

Punnett Square (Figure 2-11)

P RRYY X rryy (**cross**)
 ↓ R=round peas
 F₁ RrYy (**self**) r=wrinkled peas
 ↓ Y=yellow peas
 F₂ ? y=green peas

- 1) List the gametes from one parent on one side.
- 2) List the gametes from other parent on the other side.
- 3) Fill in gametic combinations in the squares.
- 3) Determine genetic ratios (phenotypic or genotypic) by counting squares.

Branch Diagram (Use this method!)

- 1) List probabilities for one event.
- 2) List probabilities of second event next to those of the first.
- 3) Use the product rule to determine genetic ratios.

Composition of F₂

Product rule result

	3/4 (Y-)	(3/4)(3/4) = 9/16 (R-Y-) round, yellow
3/4 (R-)	(YY,Yy,yY)	
(RR,Rr,rR)	1/4 (yy)	(3/4)(1/4) = 3/16 (R-yy) round, green
	3/4 (Y-)	(1/4)(3/4) = 3/16 (rrY-) wrinkled, yellow
1/4 (rr)	(YY,Yy,yY)	
	1/4 (yy)	(1/4)(1/4) = 1/16 (rryy) wrinkled, green

F₂ phenotypic ratio of a dihybrid cross is 9:3:3:1.

Use the product rule directly for trihybrid, etc...

F₁ AaBbCcDdEe X AaBbCcDdEe

To calculate genotypic frequency

$$p(\text{AAAbbCcDDEe}) = (1/4)(1/4)(1/2)(1/4)(1/2) = 1/256$$

To calculate phenotypic frequency

$$p(\text{A-bbC-D-E-}) = (3/4)(1/4)(3/4)(3/4)(3/4) = 81/1024$$

Pedigree Analysis

Used in human genetics (Family tree)

Symbols used in pedigree analysis (Figure 2-12).

Autosomal Recessive Disorders [Rare] (Figure 2-13)

Mendelian inheritance of an autosomal recessive disorder is revealed by the appearance of the phenotype in male and female progeny of unaffected individuals.

Inbreeding increases the chances of children being affected.

(e.g. PKU, cystic fibrosis, albinism)

Dominant disorders (Figure 2-16)

Mendelian inheritance of an autosomal dominant disorder show affected males and females in each generation, and also show affected males and females transmitting the condition to sons and daughters in equal proportions.

(e.g., achondroplasia "dwarfism")

D- (dwarfism)

dd (normal)

Interferes with bone growth during development.

(e.g., Huntington's disease)

Late-onset after having children.

Chapter 2→Patterns of Inheritance (9-9-05)

Sex Linkage→Inheritance pattern of genes on sex chromosomes.

In many organisms the gender is determined by the combination of sex chromosomes.

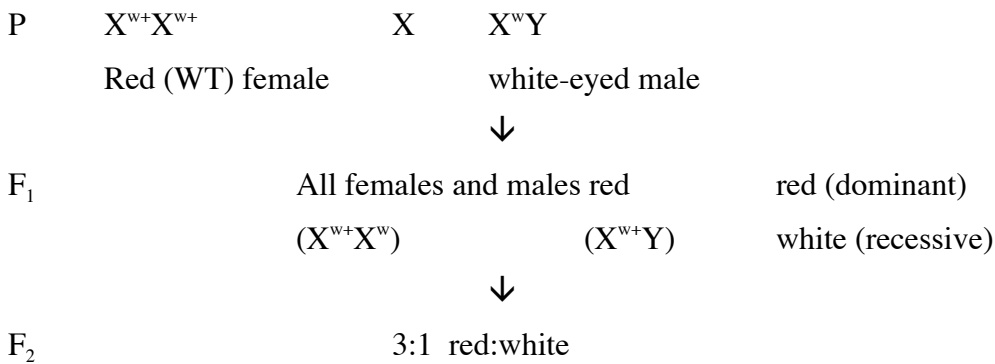
Sex chromosomes also carry genes unrelated to male and female development.

X-linked recessive inheritance→Many more males than females.

Drosophila eye color phenotype correlated with chromosome differences in microscope.

Thomas Hunt Morgan→Nobel prize (1934)

Experiment 1→Drosophila eye color (Figure 2-24)



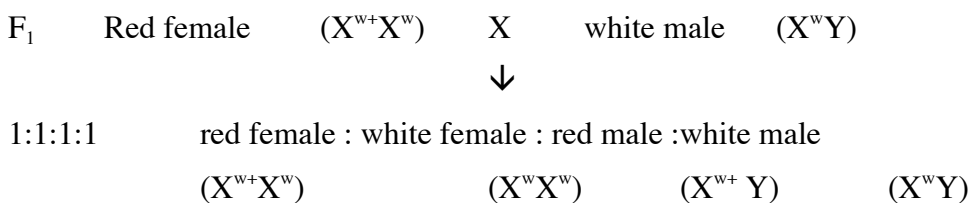
BUT:

Red: 2 female : 1 male

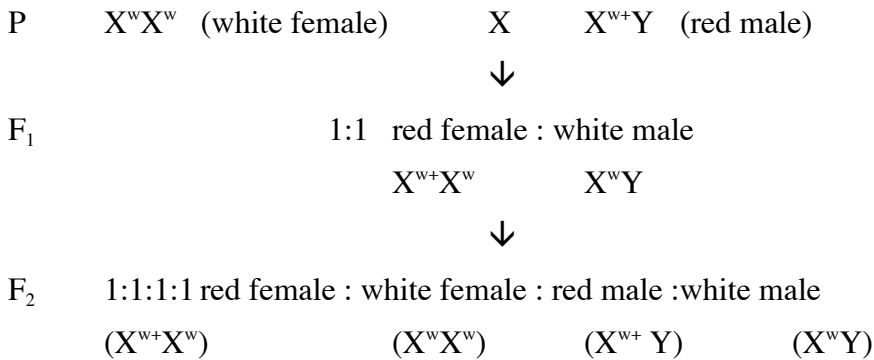
White: all male

WHY?

Experiment 2→Test Cross



Experiment 3 → Reciprocal cross of experiment 1 (Figure 2-24)

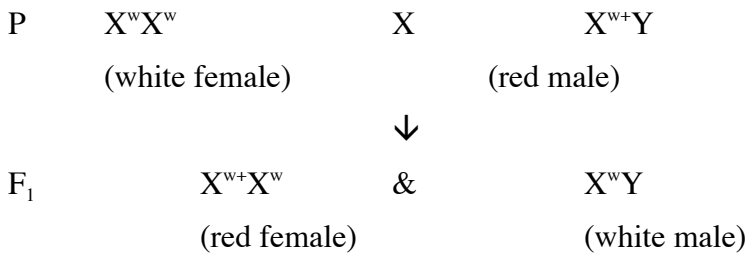


Morgan's Explanation

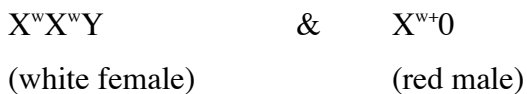
(XX) → 2 chromosomes with eye color genes.

(XY) → 1 chromosome with eye color genes.

Experiment 3 revisited



PLUS: rare exceptional progeny (1/2000)



Exceptional chromosome arrangements were observed in a microscope.

Non-Disjunction → Failure of chromosomes or sister chromatids to separate in meiosis.

Exceptional progeny arise due to non-disjunction during meiosis. (Figure 3-7)

The number of X chromosomes dictates the gender of Drosophila.

The presence of the Y chromosome dictates maleness in humans.

$X^{w+}0$ → sterile Drosophila male.

$X^{w+}0$ → sterile human female.

(Table 2-3)

Human Sex Linkage→Pedigree Analysis.

X-Linked Recessive (Figure 2-25)

Many more males than females show the recessive phenotype.

(If rare, almost all affected people are male)

If rare, none of the offspring of an affected male are affected, but all daughters are carriers.

Female carriers will pass the condition to 1/2 of her sons and 1/2 of her daughters will be carriers.

(e.g.) red-green color blindness, hemophilia

X-Linked Dominant (Figure 2-28)

Affected males pass condition to all daughters, never to sons.

Affected females pass condition to 1/2 of sons and 1/2 of daughters.

(e.g.) hypophosphatemia (Vitamin D deficient rickets)

Y-Linked Inheritance

Other than maleness, very few human phenotypes are known to be Y-linked.

X-Inactivation (p. 323)→Female mammals inherit 2 X chromosomes.

Early in development one of the 2 Xs is inactivated.

If the functional X has a recessive allele, the recessive phenotype is expressed.

Barr Body→The inactive X (microscope).

Since the inactivation process is random, all females are genetic mosaics. (i.e.) a mixture of cells having 2 genotypes corresponding to inactivation alternatives.

(e.g., sweat glands in humans; calico cats)

Chapter 3→The Nature of Chromosomes (9-12-05)

Each chromosome contains one DNA molecule.

Cytological features of Chromosomes

Chromosome number→2 to 100s depending on species.

Chromosome size→Large variation within a genome.

Centromere→Structure that attaches to spindle fibers during mitosis and meiosis. (constricted appearance at metaphase)

Metacentric→Centromere in middle

Acrocentric→Centromere off-center

Telocentric→Centromere at one end

Telomeres→Chromosome ends

Euchromatin→Active DNA (lightly packed)

Heterochromatin→Inactive DNA (tightly packed)

- A. **Constitutive**→Always inactive, permanent feature of chromosomal region
(e.g. centromeres, telomeres, inactive X chromosome)
- B. **Facultative**→Present or absent at various times
(active or inactive).

Banding Patterns→Stains result in characteristic banding patterns. (Figure 3-17a)
(e.g., G bands)

Polytene Chromosomes→Unusual feature of some flies. (Figure 3-18c)

Many rounds of replication without separation leads to a characteristic banding pattern.

Balbani rings or “puffs” are active regions of RNA synthesis (i.e., active DNA).

Chromosome Structure

E. coli → 4,600 kb → 4.6 million bp (1.3 mm)

Human cell → 6 million kb → 6 billion bp (2m)

2 m of DNA must be packaged into a 6 μm nucleus.

Human body (10^{13} cells) → sun and back 65 times; 500,000 times around earth.

Chromatin → Chromosomal DNA and protein.

Nucleosomes → DNA and chromosomal proteins called histones.

~10 nm fiber (diameter) (Figure 3-20b)

First level of packaging (beads on a string).

Each nucleosome contains an octamer of 2 subunits each of the histones H2A, H2B, H3, and H4.

DNA wrapped ~ twice around histone octamer.

Solenoid → A coil of nucleosomes.

~30 nm fiber (Figure 3-20b)

Stabilized by histone H1 that runs down the center.

Supercoil → Seen during mitosis and meiosis.

~700 nm fiber (Figures 3-22)

Held together by a scaffold at scaffold attachment regions (SARs). (Figure 3-21)

Replication & transcription of Chromatin

Nucleosomes do not dissociate.

Loosen up to allow transcription and replication.

Chromatin remodeling complexes control gene expression.

Chromosome Structure Movie

Organelle Chromosomes

Chloroplasts (cpDNA) and Mitochondria (mtDNA) contain unique genes in their own chromosome.

Inheritance patterns differ from nuclear genes.

White female x green male → all white

Green female x white male → all green

Maternal inheritance → Phenotype of all progeny is identical to the female parent.

Inheritance comes from egg cytoplasm containing chloroplasts and/or mitochondria.

Mitochondria and chloroplasts arose in eukaryotic cells by the engulfment of primitive prokaryotic cells.

Evolution led to their present function.

Mitochondria Genome (Figure 3-41a)

Mitochondria function → oxidative phosphorylation

Contain genes involved in translation and oxidative phosphorylation.

Chloroplast Genome (Figure 3-41b)

Chloroplast function → photosynthesis

Contain genes involved in transcription, translation and photosynthesis.

Requires communication between nuclear and mitochondrial/chloroplast genomes because several organelle proteins are nuclear encoded.

Communication is necessary to coordinate expression of all genes necessary for organelle function.

Nuclear encoded proteins are transported into the organelle.

Chapter 4 → Eukaryotic Linkage Mapping by Recombination (9-14-05)

Meiotic Recombination (Figures 4-6, 4-7)

Generates haploid genotypes (gametes) that differ from the haploid parental genotypes.

Interchromosomal Recombination (Figure 4-8)

If genes assort independently, crosses between a heterozygote and a tester strain generate:

50% Parental type gametes

50% Recombinant gametes

1:1:1:1 phenotypic ratio

Interchromosomal recombination movie

Gene Linkage (Figure 4-2)

Each chromosome can contain 1000s of genes.

Genes close together on the same chromosome do not assort independently.

Linked genes segregate together during meiosis.

Mendelian ratios are not observed.

Experiment 1

P $pr / pr \cdot vg / vg$ X $pr^+ / pr^+ \cdot vg^+ / vg^+$

↓

F₁ $pr^+ / pr \cdot vg^+ / vg$

$pr^+ / pr \cdot vg^+ / vg$ X $pr / pr \cdot vg / vg$

(Test Cross)

↓

pr^+	vg^+	1339	pr^+	red eye (normal)
pr	vg	1195	pr	purple eye
pr^+	vg	151	vg^+	normal wings
pr	vg^+	<u>154</u>	vg	vestigial wings
		2839		

This is not a 1:1:1:1 ratio.

Pr and vg are linked on the same chromosome.

Parental Classes→Original arrangement of alleles on the two chromosomes.

(Two most frequent classes)

Recombinant Classes→Observed less frequently than parentals.

(Figure 4-10)

Intrachromosomal Recombination

New gene combinations are generated during meiosis when non-sister chromatids cross over (X-over) between the genes under study. (Figures 4-3, cover, 4-9)

X-overs generate two reciprocal products or classes that are about equal in frequency.

Recombinant frequency (RF) significantly < 50% suggests linkage.

RF ~ 50% suggests that the genes are unlinked on separate chromosomes.

Intrachromosomal recombination movie

Linkage Mapping

To separate linked genes the X-over must occur between them.

As the distance between linked genes increase, the chance of a X-over between the genes increase.

The RF between linked genes is used to map their relative distance apart on a chromosome.

RF = 0.01 (1%) = 1 map unit (m.u.)

Gene Locus→Region on a chromosome where alleles of a certain gene are found.

Problem 1 (Revisited)

$pr^+ vg^+ / pr vg$	X	$pr vg / pr vg$	(Test Cross)
	↓		
$pr^+ vg^+ / pr vg$		1339	
$pr vg / pr vg$		1195	
$pr^+ vg / pr vg$		151	
$pr vg^+ / pr vg$		<u>154</u>	
		2839	

305 recombinant x 100 = 10.7% = 10.7 m.u.

2839 total

10.7% < 50% pr and vg are linked

Problem 2

Given a genetic distance in m.u., we can predict frequencies of progeny from a test cross.

$a^+ b^+ / a b$ X $a b / a b$ (Test Cross)

If 20 m.u. apart:

$a^+ b^+ / a b$	40%
$a b / a b$	40%
$a b^+ / a b$	10%
$a^+ b / a b$	10%

Chapter 4 → Eukaryotic Chromosome Mapping by Recombination (9-16-05)

Three-Point Testcross

Map units are additive, but to order 3 genes we need to perform a three-point testcross. (Figure 4-11)

Double X-overs → Smallest class because two X-overs required.

Compare parental types with double X-overs. The gene that is "switched" is in the middle.
(Figures 4-12 and 4-13)

P	$v^+ / v^+ \cdot cv / cv \cdot ct / ct$	X	$v / v \cdot cv^+ / cv^+ \cdot ct^+ / ct^+$	
	↓			
F ₁	$v / v^+ \cdot cv / cv^+ \cdot ct / ct^+$		(Test Cross)	
	↓			
F ₂	v	cv ⁺	ct ⁺	580
	v ⁺	cv	ct	592
	v	cv	ct ⁺	45
	v ⁺	cv ⁺	ct	40
	v	cv	ct	89
	v ⁺	cv ⁺	ct ⁺	94
	v	cv ⁺	ct	3
	v ⁺	cv	ct ⁺	<u>5</u>
				1448

- 1) Consider v and cv (neglect ct for now)
v ct⁺ & v⁺ ct parentals; v ct & v⁺ ct⁺ recombinants
RF = (89 + 94 + 3 + 5) / 1448 = 0.132 = 13.2%

- 2) Now consider v and ct (neglect cv)
cv⁺ ct⁺ & cv ct parentals; cv⁺ ct & cv ct⁺ recombinants
RF = (45 + 40 + 3 + 5) / 1448 = 0.064 = 6.4%

- 3) Now consider cv and ct (neglect v)
v cv⁺ & v⁺ cv parentals; v cv & v⁺ cv⁺ recombinants
RF = (45 + 40 + 89 + 94 + 3 + 3 + 5 + 5) / 1448 = 0.196 = 19.6%

Now construct the linkage map

v 13.2 ct 6.4 cv

Interference

A X-over in one region of the chromosome decreases the chances that a X-over will occur in an adjacent region.

Calculate the frequency and number of double recombinants expected if there is no interference.
(i.e., use the product rule)

$$\text{Expected frequency} = (0.132)(0.064) = 0.0084$$

$$\text{Expected number} = (0.0084)(1448) = 12$$

Interference (I) = 1 - c.o.c. (coefficient of coincidence)

$$I = 1 - \frac{\text{observed number of double X-overs}}{\text{expected number of double X-overs}}$$

$$I = 1 - 8/12 = 4/12 = 1/3 = 0.33 = 33\%$$

This is another example of a three-point testcross.

$a^+ a b^+ b c^+ c$	X	$a a b b c c$
		↓
$a^+ b^+ c$	788	
$a b c^+$	802	
$a^+ b c$	72	
$a b^+ c^+$	68	
$a^+ b^+ c^+$	128	
$a b c$	122	
$a b^+ c$	9	
$a^+ b c^+$	<u>11</u>	
	2000	

Construct a linkage map and calculate interference.

First determine the middle gene a

- 1) Consider a and b (neglect c)
 $a^+ b^+, a b \rightarrow$ parentals; $a^+ b, a b^+ \rightarrow$ recombinants
 $RF = (72 + 68 + 9 + 11) / 2000 = 160 / 2000 = 0.08 = 8\%$

- 2) Consider a and c (neglect b)
 $a^+ c, a c^+ \rightarrow$ parentals; $a^+ c^+, a c \rightarrow$ recombinants
 $RF = (128 + 122 + 9 + 11) / 2000 = 270 / 2000 = 0.135 = 13.5\%$

- 3) Consider b and c (neglect a)
 $b^+ c, b c^+ \rightarrow$ parentals; $b^+ c^+, b c \rightarrow$ recombinants
 $RF = (72 + 68 + 128 + 122 + 9 + 9 + 11 + 11) / 2000 = 430 / 2000 = 0.215 = 21.5\%$

c 13.5 a 68.0 b

$$I = 1 - (O/E) \quad E = (0.08)(0.135)(2000) = 21.6$$

$$I = 1 - (20/21.6) = 1 - 0.926 = 0.074 = 7.4\%$$

Chapter 5→Bacterial and Bacteriophage Genetics (9-19-05)

Prokaryotes→Eubacteria and Archaeobacteria

Divide by binary fission resulting in genetically identical progeny. (Figure 5-2)

Single, circular chromosome.

No mitosis, meiosis, nucleus.

Haploid.

Bacteriophage (Phage)→bacterial virus.

Not free living→parasitize bacteria to replicate.

Conjugation→Transfer of DNA from one cell to another via direct cell-to-cell contact. (Figure 5-7)

Requires the fertility (F) factor→small, circular DNA element that can replicate (mini chromosome).

Cells with F are F⁺; cells without F are F⁻.

F⁺ donor cells produce pili which attach to F⁻ recipient cells.

Donor transfers a copy of F to the recipient.

A copy of F always remains in donor (i.e., replication).

F can integrate into the chromosome by X-ing over, thereby generating an Hfr strain. (Figure 5-8)

Hfr Strains (high frequency of recombination)

Can transfer chromosomal genes to F⁻ recipient.

DNA fragment can recombine with chromosome and generate recombinants. (Figure 5-10)

Conjugation Movie

Determining Linkage (Interrupted Mating)

(Figure 5-11)

Donor Recipient
Hfr (a⁺ b⁺ c⁺ d⁺ str^S) X F⁻ (a⁻ b⁻ c⁻ d⁻ str^R)

Time course, remove and plate samples on rich medium containing streptomycin (str), an antibiotic that prevents survival of donor cells.

Screen surviving cells for transferred genes by replica plating.

Origin → Fixed point on donor chromosome where gene-transfer starts (Hfr transferred last).

Orientation of integrated F determines polarity (direction) of transfer.

First gene transferred depends on orientation and position of F.
(origin → terminus)

Many Hfr strains exist depending on position & orientation of F. (Figure 5-13)

Bacterial Mapping (*E. coli*)

Bacterial matings result in a partial diploid (merodiploid).

Transferred genes must recombine with recipient chromosome.

Results in exchange of genetic material → requires double X-over (Not a reciprocal change)
(Figure 5-15)

(Figure 5-14)

Determining gene order from interrupted mating

Hfr leu⁺ met⁺ arg⁺ str^S X F⁻ leu⁻ met⁻ arg⁻ str^R

1. Determine the gene that is closest to the origin by performing an interrupted mating experiment (met⁺).
2. Then with a natural gradient of transfer experiment plate on minimal + all supplements except methionine.
(i.e. select for met⁺ recombinants)
3. Screen for other genes by replica plating.

100% met⁺
60% arg⁺
20% leu⁺

Gene order → leu arg met
----->

High Resolution Mapping (Figure 5-16)

Hfr leu⁺ arg⁺ met⁺ str^S X F⁻ leu⁻ arg⁻ met⁻ str^R
-----> -----

1. select for last gene to enter (i.e., leu⁺)
Thus, every transferred fragment has all 3 genes.
2. screen for the other genes by replica plating

1 map unit = 1% X-over in the interval

leu ⁺ arg ⁻ met ⁻	8	(8/200 = 4% = 4 map units)
leu ⁺ arg ⁺ met ⁻	18	(18/200 = 9% = 9 map units)
leu ⁺ arg ⁺ met ⁺	<u>174</u>	(174/200 = 87%)
	200	

And, rare quadruple X-overs leu⁺ arg⁻ met⁺
(determines the middle gene)

leu	arg	met
4	9	

Product rule (0.05)(0.05)=0.0025=0.25%

Bacterial Transformation → Conversion of one genotype to another by introducing chromosomal DNA.
Requires recombination (similar to Hfr x F⁻ cross).

If two genes are close to each other both can be transferred on the same DNA fragment.
Limited use for genetic mapping.

Bacteriophages

Virulent Phages → Always lytic

Temperate Phages → Lytic or lysogenic

Lysogeny → Phage infected bacteria not leading to lysis

Lysogenic strains carry a prophage (phage DNA with lytic functions turned off).

Can be integrated or as a separate circle.

UV light or chemicals induce prophage to enter the lytic cycle.

Transduction

Phage can transfer bacterial genes from one cell to another by packaging bacterial DNA by mistake.

Generalized Transducing Phage

Can carry any part of the bacterial chromosome.

Specialized Transducing Phage

Can only carry a specific chromosomal region.

Generalized Transduction

When a cell is infected by a non-lysogenic phage the bacterial chromosome is broken up.

Phage can package pieces of bacterial DNA instead of phage DNA.

(e.g.) P1 (Figure 5-27)

Transducing phage ejects bacterial donor DNA into a recipient strain → merodiploid → recombination.

Linkage data from P1 transduction

Genes must be close enough for P1 to transduce them in a single piece of DNA.

- 1) (Donor) P1 $\text{met}^+ \text{arg}^+$ X $\text{met}^- \text{arg}^-$ (Recipient)
- 2) Select for met^+ (plate on minimal with arginine)
- 3) Screen for the % of met^+ that are also arg^+ .

Strains transduced to $\text{met}^+ \text{arg}^+$ are cotransductants.

The greater the cotransduction frequency the closer the genes are to each other.

(Donor) P1 $\text{a}^+ \text{b}^+ \text{c}^-$ X $\text{a}^- \text{b}^- \text{c}^+$ recipient

Select for a^+ then screen for $\text{b}^{+/-}$ and $\text{c}^{+/-}$.

<u>Genotype</u>	<u># Colonies</u>
$\text{a}^+ \text{b}^+ \text{c}^+$	7
$\text{a}^+ \text{b}^+ \text{c}^-$	20
$\text{a}^+ \text{b}^- \text{c}^+$	46
$\text{a}^+ \text{b}^- \text{c}^-$	27
	<hr/>
	100

The cotransduction frequency of a and b is 27%

The cotransduction frequency of a and c is 47%

Specialized Transduction

(e.g., bacteriophage lambda (λ))

Integration by a single X-over between gal and bio.

Useful for moving specific genes. (i.e.) gal or bio

(Figures 5-30, 5-31a)

Mapping genes on *E. coli* Chromosome

- 1) First use Hfr strains to map genes within about 10-15 minutes (min).
- 2) Then use P1 transduction within the interval established by Hfr conjugation.

Conjugation, transformation and transduction require an equal number of X-overs to replace DNA in the bacterial chromosome.

Chapter 6→From Gene to Phenotype (9-23-05)

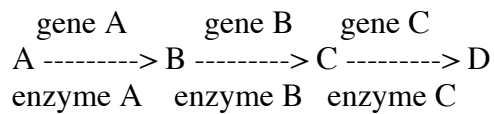
Background Information

Genes specify the structure of proteins.

Genes do not act in isolation. (i.e. the gene products (protein) interact with one another).

Different alleles of a gene specify different forms of the corresponding protein. (e.g. active, inactive, etc.)

Cellular processes occur by pathways in which enzymes act in a series.



Incomplete Dominance

Heterozygotes have intermediate phenotypes of the two homozygotes.

(e.g., plant flower color)

P	AA	X	aa	(Cross)
	Red		White	
		↓		
F ₁	Aa	(Self)		
	Pink			
		↓		
F ₂	1 AA	Red		
	2 Aa		Pink	
	1 aa		White	

Codominance→Heterozygote has phenotype of both homozygotes.

(e.g. Human Hemoglobin)

Hb ^A Hb ^A	Normal Red Blood Cells (RBCs).
Hb ^S Hb ^S	Abnormal “sickle” shaped RBCs, anemia (often fatal).
Hb ^A Hb ^S	No anemia, RBCs form sickles at low [oxygen].

- 1) Regarding anemia, Hb^A is dominant.
- 2) Regarding cell shape, Hb^A is incompletely dominant.
- 3) Regarding hemoglobin molecules, Hb^A and Hb^S are codominant.

Multiple Alleles

A gene with more than two alleles.

(e.g.) Human ABO Blood Groups (3 alleles)

<u>Genotype</u>	<u>Blood Group</u>
$I^A / I^A, I^A / i$	A
$I^B / I^B, I^B / i$	B
I^A / I^B	AB
i / i	O

I^A and I^B are fully dominant in $I^A i$ and $I^B i$ and codominant in $I^A I^B$.

AB → Universal acceptor (both A and B Ag on cell surface)

O → Universal donor (no antigen on cell surface)

Lethal Alleles

Alleles of a gene that cause lethality. May be lethal if homozygous or even heterozygous.

Lethality may occur early during embryonic development, after birth, or later in life.

Many human lethal genes result in spontaneous abortions.

Mouse coat color

Dark coat → Normal (W.T.); Yellow coat → Variant

P Dark (AA) X Yellow ($A^Y A$)

↓

F_1 1:1 Dark:Yellow

P Yellow ($A^Y A$) X Yellow ($A^Y A$)

↓

F_1 2:1 Yellow:Dark (Suggests no homozygous yellow).

1/4 AA (Dark)

1/2 $A^Y A$ (Yellow)

1/4 $A^Y A^Y$ (Die Before Birth)

Pleiotropic

Allele that causes more than one phenotype.

(e.g., mouse coat color. Yellow when heterozygous, lethal when homozygous).

(e.g., manx cats. Tailless when heterozygous, lethal when homozygous).

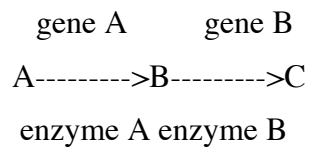
(e.g., human blood clotting mutations)

Complementation

Wild type phenotype requires at least one dominant allele of each of two genes.

Homozygosity for the recessive allele of either gene results in the recessive phenotype.

(e.g.) Pea petal color (biochemical pathway)



P	(AA bb) white line 1	X	white line 2 ($aaBB$)
			↓
F ₁			AaBb (purple)
			↓
F ₂		9	A-B- (purple)
		3	A-bb (white)
		3	aaB- (white)
		1	aabb (white)

9:7 F₂ ratio if different genes; all recessive if the same gene.

Epistasis

The ability of a mutation at one locus to override a mutation at another in a double mutant.

Overriding mutation is epistatic, overridden mutation is hypostatic.

	gene A		gene B
	white ----->	magenta ----->	blue
	enzyme A		enzyme B
P	aaBB (white)	X	AAbb (magenta)
		↓	
F ₁	AaBb (blue)		
		↓	
F ₂	9	A-B- (blue)	
	3	A-bb (magenta)	
	3	aaB- (white)	
	1	aabb (white)	

9:3:4 F₂ ratio if gene A is epistatic to gene B

Suppressors

(e.g., an enzyme consisting of two separate polypeptides)

bb = second site suppressor that suppresses the mutant aa phenotype

P	aaBB (purple)	X	AAbb (red)
		↓	
F ₁	AaBb (red)		
		↓	
F ₂	9	A-B- (red)	
	3	A-bb (red)	
	3	aaB- (purple)	
	1	aabb (red)	

13:3 F₂ ratio if gene s suppresses the mutant a phenotype