Office hours:
Monday 5-6pm (door closes @ 5:15p)
Friday 4-5 pm
11 Koshland Hall

What does a geneticist mean by “gene”? …revealed by how a geneticist tests for “allelism”
Alleles: DIFFERENT (alternative) forms of the same gene

Given:
- mutant a (lozenge-shaped eyes) vs. wildtype
- mutant b (lozenge-shaped eyes) vs. wildtype

Are mutant A and mutant B alleles?
YES: A&B are alternative forms of the same gene
NO: A&B are alternative forms of different genes

Alleles:
DIFFERENT (alternative) forms of the same gene

What does a geneticist mean by “gene”?… revealed by how a geneticist tests for “allelism”

Given:
- mutant a vs. wildtype
- mutant b vs. wildtype

Before we ask:
Are mutant a and mutant b alleles?

First must ask:
(1) are each of these mutant phenotypes likely to be caused by single-gene differences from the wildtype?
(2) do the mutants map to the same general region of the genome?

(sequencing mutant A and mutant B genomes would not necessarily answer either question!)

Given:
- mutant a vs. wildtype
- mutant b vs. wildtype

(1) are each of these mutant phenotypes likely to be caused by a single-gene differences from the wildtype?
(2) do the mutants map to the same general region of the genome?

low resolution segregation tests:
(1) a/a X +/+ → a/+ → a/a & a/+ & +/+ only
(2) m1 m2; a/+ X m1 m2; a/+ → trihybrid → least frequent gamete classes:

3-factor cross rule: least frequent class:
two outside parental, inside nonparental:
hence order: m1 a m2

Are a & b alternative units of segregation during meiosis?
Are they segregational alleles?

Do we go with Mendel or (Lewis)Benzer?

high-resolution segregation test for recombination
breeding behavior of hybrids

complementation test for function
phenotype of hybrids

mutant a

mutant b

a + b

OR

a

b

Are a & b alternative units of segregation during meiosis?
Are they segregational alleles?

i.e. are they segregational alleles?

complementation test for function
phenotype of hybrids

mutant a

mutant b

a + b

OR

a

b

must first establish:
that a & b are recessive?

Only

high-resolution segregation test for recombination
breeding behavior of hybrids

mutant a

mutant b

a + b

OR

a

b

wildtype

Phenotype

Phenotype

(Complement)

(hence one functional copy sufficient)

OR

a

b

wildtype

Phenotype

Phenotype

(Complement)

(hence one functional copy sufficient)

complementation test for function
phenotype of hybrids

mutant a

mutant b

a + b

OR

a

b

are they functional alleles?

are they functional alleles?

high resolution
segregation test
for
recombination
breeding behavior of hybrids

complementation test for function
phenotype of hybrids

mutant a

mutant b

a + b

OR

a

b

are they functional alleles?

are they functional alleles?

Phenotype

Phenotype

(Complement)

(hence one functional copy sufficient)
Are mutant a and mutant b alleles (i.e. genetic alternatives)?

What about the cis/trans test?

Where is the cis control

mutant a
 wildtype = w.t. pheno.
mutant b
 wildtype = w.t. pheno.

a & b in cis

mutant a mutant b

Ik can believe it!

Are mutant a and mutant b alleles (i.e. genetic alternatives)?

--- are we being misled by the apparent failure of these recessive mutants to complement in trans?

When do we need the cis control

mutant a mutant b

...could mutants be recessive individually and in different cis-acting units of genetic function (i.e. not be functional alleles, but interact in combination to appear dominant together).

If so, the cis control will also be mutant.

Are mutant a and mutant b alleles (i.e. genetic alternatives)?

CIS control

mutant a mutant b

wildtype phenotype...
we can believe it!

mutant a mutant b

wildtype phenotype

...we've been misled!

mutant a mutant b

wildtype phenotype

(complement) (don't complement)

not alleles alleles

But if mutant phenotype

cis vs. trans matters!

CIS phenotype ≠ TRANS phenotype, hence functional alleles

BUT if mutant phenotype

cis vs. trans doesn't matter!

CIS phenotype = TRANS phenotype, hence NOT functional alleles

Are mutant a and mutant b alleles (i.e. genetic alternatives)?

CIS

mutant a mutant b

mutant a mutant b

The true cis/trans test will allow us to determine allelism even if one or both of the mutants are not recessive!

CIS phenotype ≠ TRANS phenotype, hence functional alleles

CIS phenotype = TRANS phenotype, hence NOT functional alleles

...and all that matters is whether the cis vs. trans phenotypes are the same or different, not whether either one is wildtype.

Remember: the "complementation test" per se is limited to recessive mutants. Most mutants are recessive, but some of the most useful & interesting are not.
Are mutant a and mutant b alleles (i.e., genetic alternatives)?

- Complementation test for function alleles as alternative cis-acting units of function
- Conflict favored
- High-resolution segregation test for recombination: alleles as alternative units of segregation (recombination)

Intuitively, a better operational definition of alternative forms of genes

**Genetic map** (linear like chromosome)

The basis for making genetic maps

**Rearranges questions:**

If one can get recombination between functional alleles (alternative forms of a gene), then how do the genetic maps one can therefore construct within single genes compare to the genetic maps that can be (had been) constructed between genes?

--- **WHAT IS THE NATURE OF GENETIC FINE STRUCTURE?**

...and what is the basic (minimal) unit of recombination (i.e., what are true segregational alleles)?

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**American Naturalist, v56 p32 (1922)**

**VARIATION DUE TO CHANGE IN THE INDIVIDUAL GENES**

DR. H. J. MULLER

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That two distinct kinds of substances—**the different substances and the genes**—should both possess this most remarkable property of heritable variation or **mutation**. A germ working as a single, different mechanism, is quite conceivable; considering the complexity of proteoplasm. Yet it would seem a curious coincidence indeed. It would open up the possibility of two totally different kinds of life, working by different mechanisms. On the other hand, if these different bodies were really genes, fundamentally like the chromosomes, genes that would give us an entirely new angle from which to view the gene problem. They are heritable, as some exterms tell, can be analyzed in test-tubes, and their properties, as shown by their effects on the bacteria, can then be studied after treatment. It would be very nice to call these bodies genes, and yet at present we must confess that there is no distinction known between the genes and chromosomes.

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**Fig. 7.20** p228

**Phage kill bacteria for a living (and replicate in the process). geneticists count “centers of killing” (plaques)**

**Presence indicated as plaques**

- add: 100 phage in 0.1 ml media to: 10^9 cells in 0.1 ml media
- let sit 20 min
- add: 2.5 ml molten agar
- pour on a plate
- grow overnight at 37° C

mol is 100:10 = 1:100,000...

.....the chance of a single cell being infected by 2 phage is 10^-10

“**multiplicity of infection**”

---

**T4 plaques on a “lam” of E coli cells**

**Must have a phenotypic difference between pure-breeding lines to do genetics**
Phage phenotypes (differences): based on growth

- plaque morphology: large/small (growth rate), clear/turbid, smooth edged/rough edged
- growth conditions: bacterial host range, temperature range (T4: 25-42°C)

By early 1950s, Alfred Hershy had isolated a number of phage T4 mutants and constructed a (circular) genetic map based on:

1. complementation (to group the mutants into genes)
2. recombination to organize those genes on a map.

...and along came physicist Seymour Benzer

The young S. Benzer: (1950s)

- phage T4 as his genetic workhorse

An older Benzer: (1967-present)

- fruit fly (D. melanogaster) as his genetic workhorse

- reconciled the segregational and functional definitions of the gene, set up the experimental system used in the 2nd most elegant genetics paper in history
- founded many of the most interesting areas of modern behavioral genetics (clocks & learning, etc.)
- set up the experimental system most effective for studying fly development: the compound eye

P.N.A.S.-US v41p344 (1955)

FINE STRUCTURE OF A GENETIC REGION IN BACTERIOPHAGE
By Seymour Benzer

This paper describes a functionally related region in the genetic material of a bacteriophage that is indivisible by mutation and by genetic recombination. The region of mutants resembles similar cases which have been observed in many organisms, usually designated as “pseudo alleles.” (See reviews by Lewin and Pentecost.) Such cases are of special interest for their bearing on the structure and function of genetic determinants.

The phenomenon of genetic recombination provides a powerful tool for separating mutations and discerning their positions along a chromosome. When it comes to very closely neighboring mutations, a difficulty arises, since the closer two mutations are to one another, the smaller is the probability that recombination between them will occur. Therefore, failure to observe recombinant types among a finite number of progeny ordinarily does not justify the conclusion that the two mutations are inseparable but can only place an upper limit on the linkage distance between them. A high degree of resolution requires the examination of very many progeny. This can best be achieved if there is available a selective feature for the detection of small proportions of recombinants.