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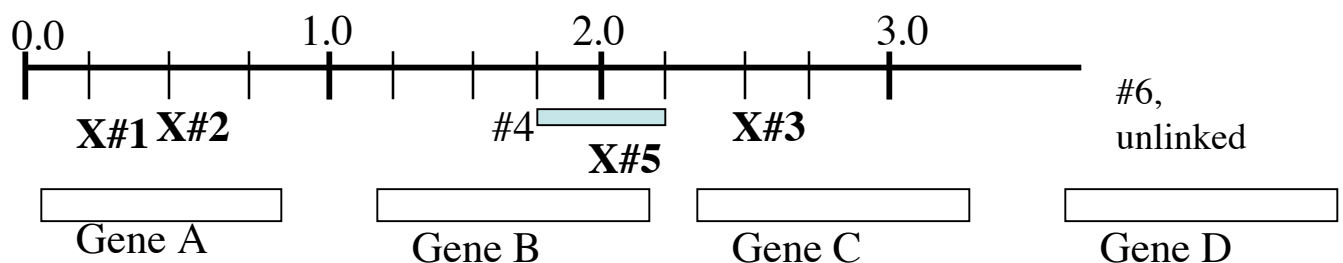
Problem Set 3 Genetics 320 Due Sept 12

1. Recombination and complementation in phage
 - A. Describe how you would do a recombination and complementation test, given you have permissive and restrictive host cells for phage yabadabdo. Be succinct.

Perform a recombination test with each pair of mutants, using the permissive host to allow recombination, then test for formation of wt phage by plating on the restrictive host. The # of wt x 2 over the total phage plated (determined by plating on permissive host) gives the frequency of recombination.

Complementation is performed by co-infecting the restrictive host and looking for plaques.
 - B. Below are the data you generated in analysis of the mutants. Mutant #1 is given with an "X". On the map below, place where the other 5 mutations lie. If one of the mutations is a deletion, draw a bar to indicate the region deleted (note bar for mutation #7 as an example. >50% recombination means the mutations are not linked..
 - C. Which mutations are recessive and which recessive. *All recessive*
 - D. Indicate the location of the genes consistent with the data (label genes A, B etc)

Cross	Complementation	Recombination (%)
1x1	-	0
1x2	-	0.25
1x3	+	2.25
1x4	+	1.5
1x5	+	1.75
1x6	+	52
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2x3	+	2.0
2x4	+	1.25
2x5	+	1.5
2x6	+	51
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3x4	+	0.25
3x5	+	0.5
3x6	+	53
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4x5	-	0
4x6	+	51
5x6	+	51

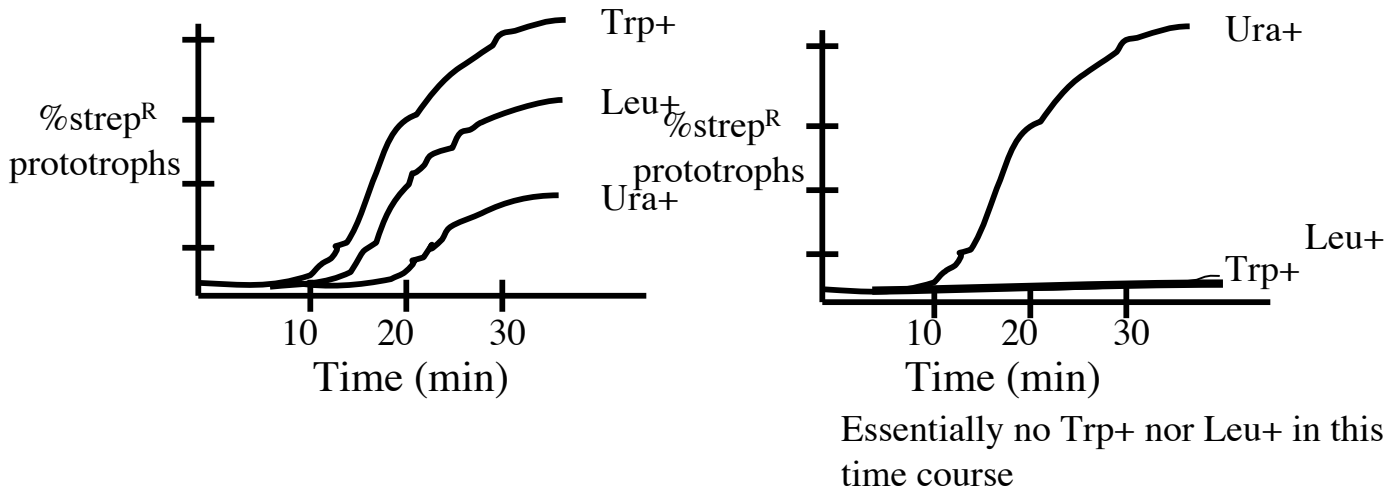


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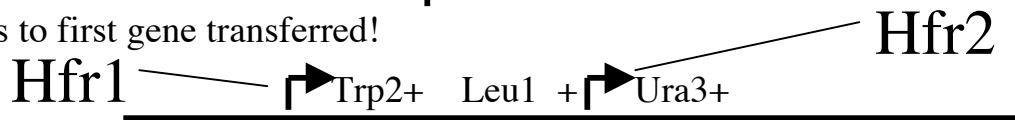
2. An Hfr cross is performed with the following two Hfr strains (Hfr1 and Hfr2)

A. Hfr1 Leu1+ Trp2+ Ura3+ Strp^S by F- Leu1- Trp2- Ura3- Strp^R

B. Hfr2 Leu1+ Trp2+ Ura3+ Strp^S by F- Leu1- Trp2- Ura3- Strp^R



From these data, place n the line below where each of the two Hfr are located and their direction of transfer. Use this to indicate direction- arrow points to first gene transferred!



3. Here is another Hfr cross and data.

Hfr3 Bio1+ Ade2+ Arg2+strepR x F-bio1- Ade2- Arg2- StrepS

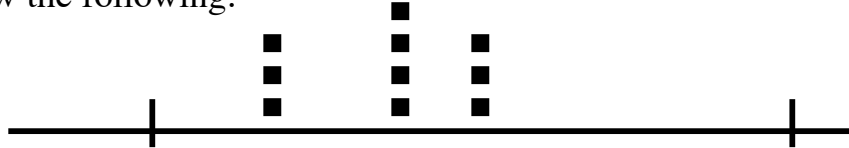
F- recipient	Frequencies of transfer
Bio1+ Ade2+ Arg2+	620
Bio1+ Ade2+ Arg2-	20
Bio1+ Ade2- Arg2+	300
Bio1+ Ade2- Arg2-	270

Indicate the likely gene order.

Bio1+ Arg2+ Ade2+

4. Crick-Brenner test of the genetic code

A. A new scientist tried to repeat the phage experiments. He used a mutagen, like EMS, that alkylates bases, and therefore changes base-pairing. When plated on B, he got mostly fuzzy roundish plaques that did not form plaques on K. He mapped the mutations and saw the following:



i. Provide 1 reason why there are “hotspots” of mutation.

Those particular DNA sequences could be prone to base changes. More likely, the amino acids in that region alter protein function when changed.

ii. There were some round plaque. These mutations were never suppressed by proflavin-induced mutations. Explain.

These mutations probably introduced stop codons into the gene, but not by frameshift, rather by base-pair change-or at least a drastic change in protein function. The key is that a frameshift mutation (induced by proflavin) would not restore activity, and would actually itself introduce a frameshift and further destroy the protein.

iii. Fuzzy mutations were also not suppressed by proflavin-induced mutations. Explain.

These are base pair changes that alter the aminoacid, but do not alter reading frame. As for ii above, a frameshift mutation added to a basepair change does not restore reading frame.

5. The original rII-1 proflavin-induced mutation was used to isolate suppressors, which were then separated by recombination from rII-1. The suppressors were used to isolate suppressors as well. Each suppressor was given a “sign”.

rII-1 +

rII-2 -

rII-73 -

rII-46 -

rII-19 +

rII-23 +

i. A new mutation, rII-?, was isolated as a suppressor of rII-73. How would you determine if this is a new mutation or is identical to another mutation on this list? *Do a recombination test with each of the known mutations. If no wt are recovered from one of them, then the new mutation is the same (or extremely close!) to the one known mutation.*

ii. List 2 sets of 3 mutations that should show suppression, and 2 sets that will not.

rII-1, -19, -23 & rII-2, 73, 46 show suppression

rII-1, -19, 2 & rII-19, 73, 2 don't show suppression (and many many others too!)