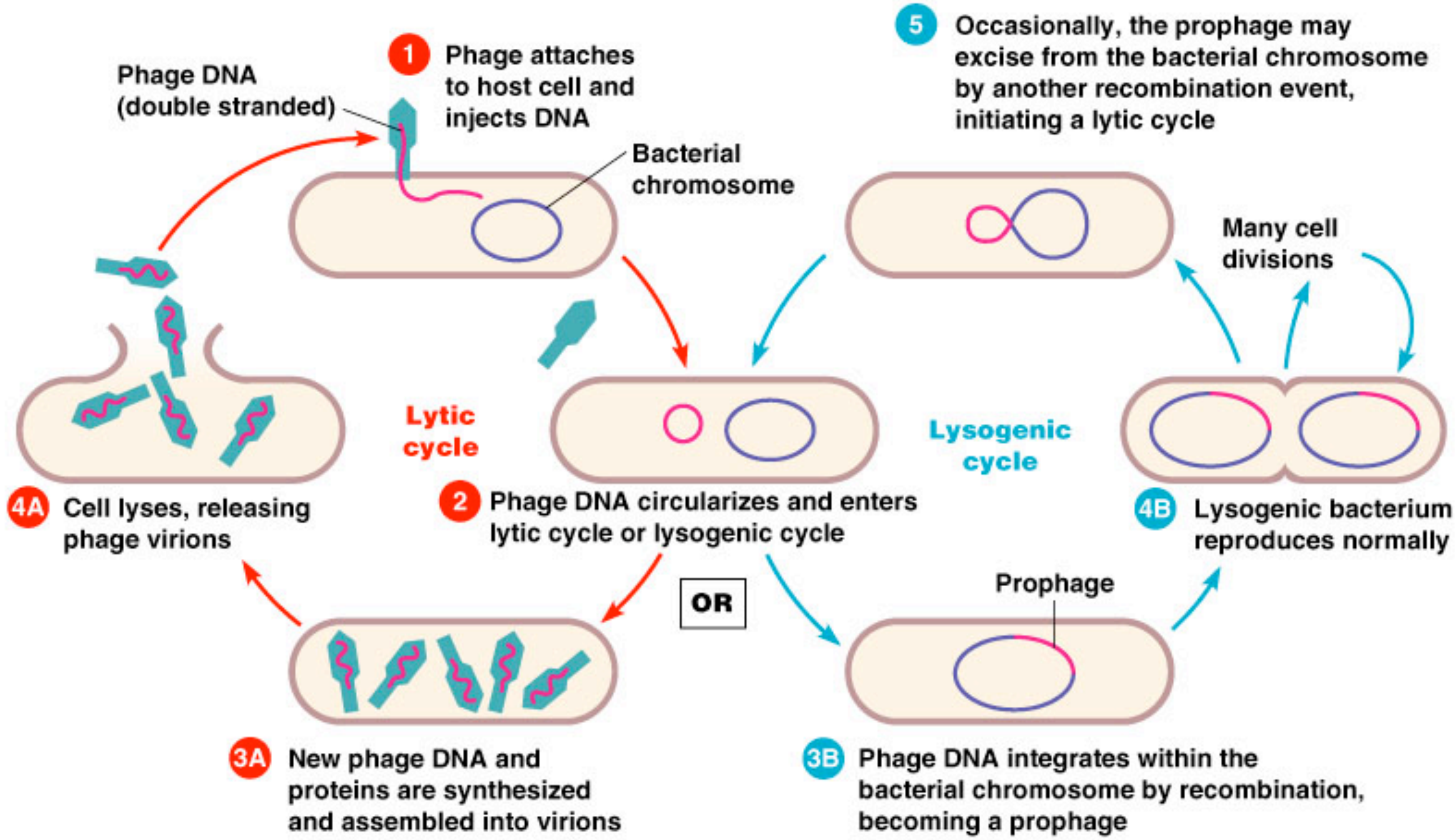
Chapters 7 and 8: Transduction

Transduction:

Bacteriophages: viruses that infect bacteria

- **Obligate parasite: must use host machinery for producing more phage particles**
- **dsDNA, ssDNA, or RNA encapsulated by a protein or membrane coat**
  - **Capsid:**
- **Complex tail structure for injecting DNA through the cell membrane**

2 Cycles for Viral Particle Production: Lytic versus Lysogenic Phage

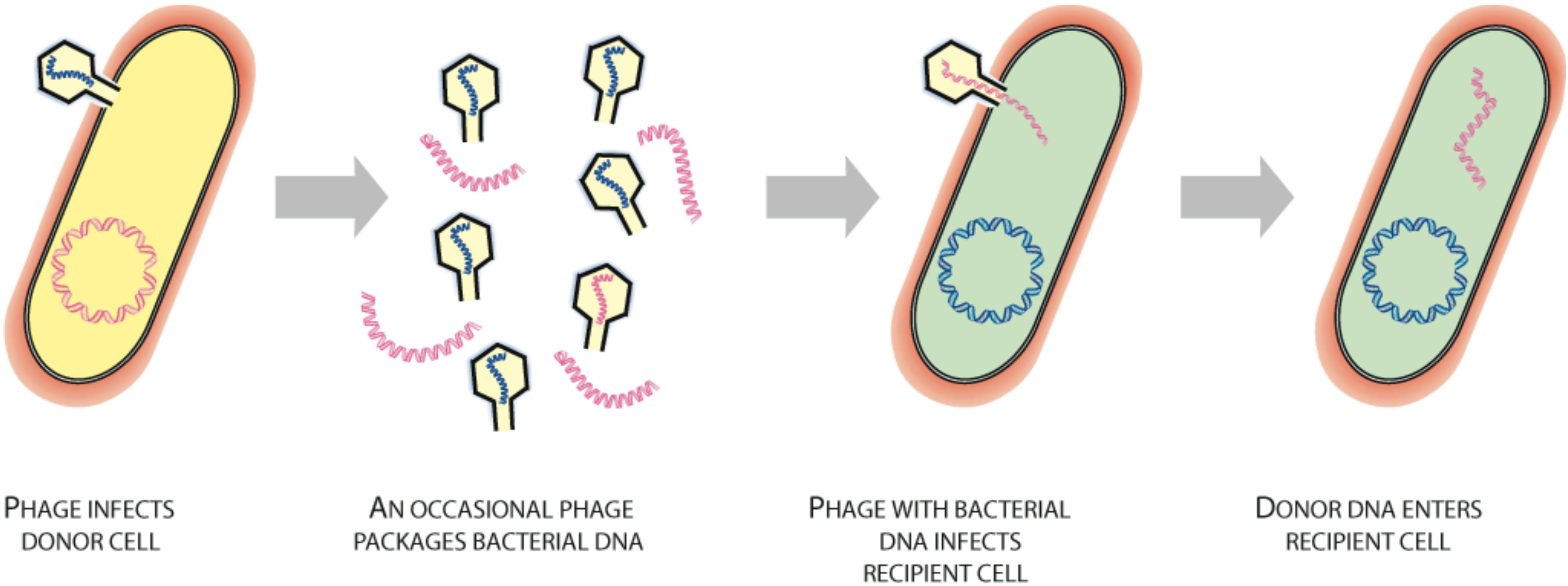


Close Up of Lytic Cycle and Lysogeny

Virulent phage vs. Temperate phage

Fig. 7.2 and 8.1

Process of Transduction



1)

2)

3) During amplification of phage particles, bacterial DNA can sometimes be packaged instead of viral DNA

4) Once donor/host lyses, transducing particle is free to infect another recipient cell and inject its DNA

2 Types of Transduction

A) Specialized Transduction-

1)

2) Only host genome regions that flank the prophage are packaged by mistakes in recombination

3) A single transducing particle can be used to make many identical particles through phage reproduction

4)

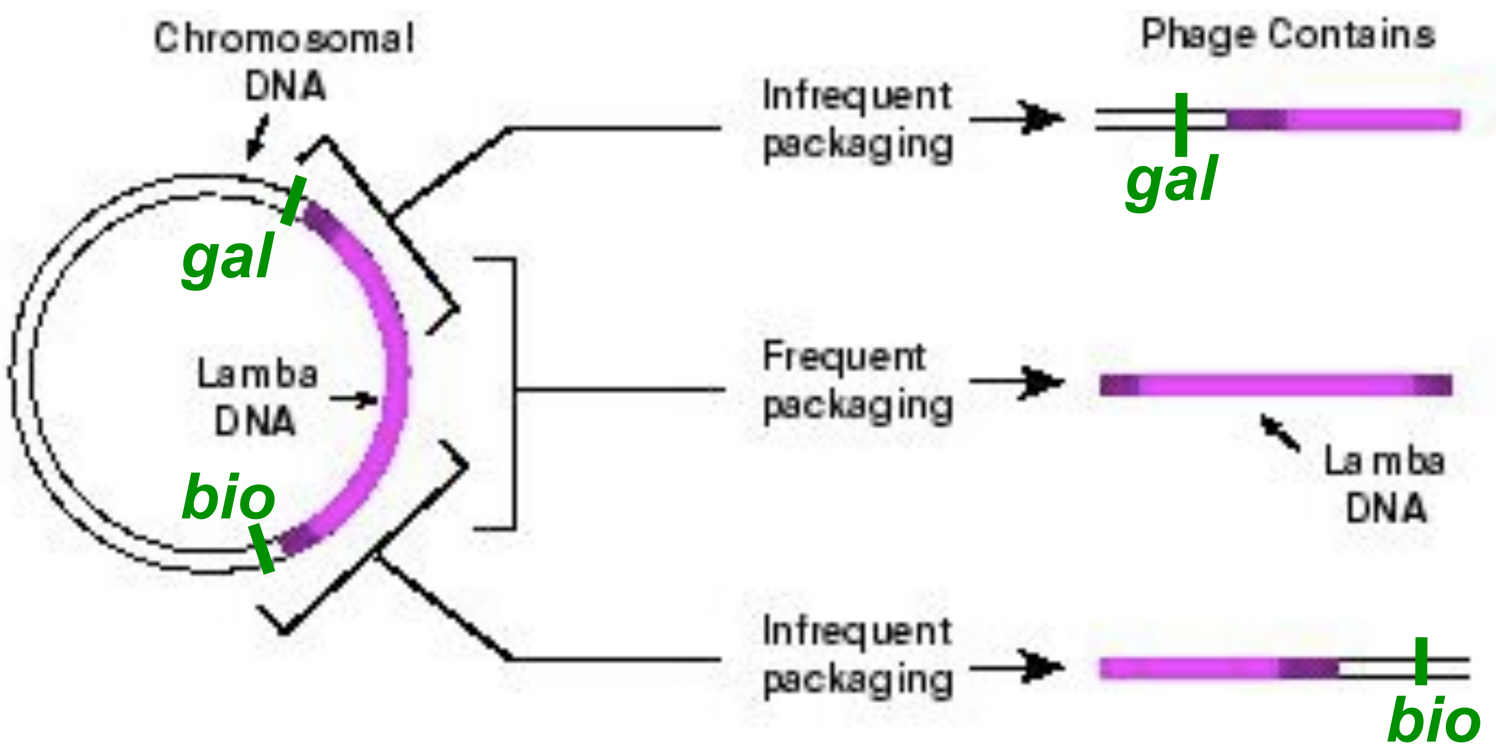
Specialized transduction by λ (lambda)

λ DNA inserts at the *attB* site within the *E. coli* genome

E. coli **chromosome** ***attB*** **locus:** ***gal - attB - bio***

Organization in lysogenic ***E. coli*** **:**

gal - att - int - N - cI - cos - **b2 region** ***- att - bio***



Low-Frequency Transducing (LFT) Lysate:

•

• Results in either a λd*gal* or a λd*bio* transducing particle

- d: stands for defective phage; improper excision generally results in some of the phage DNA being left in the host's chromosome

• Very rare in occurrence

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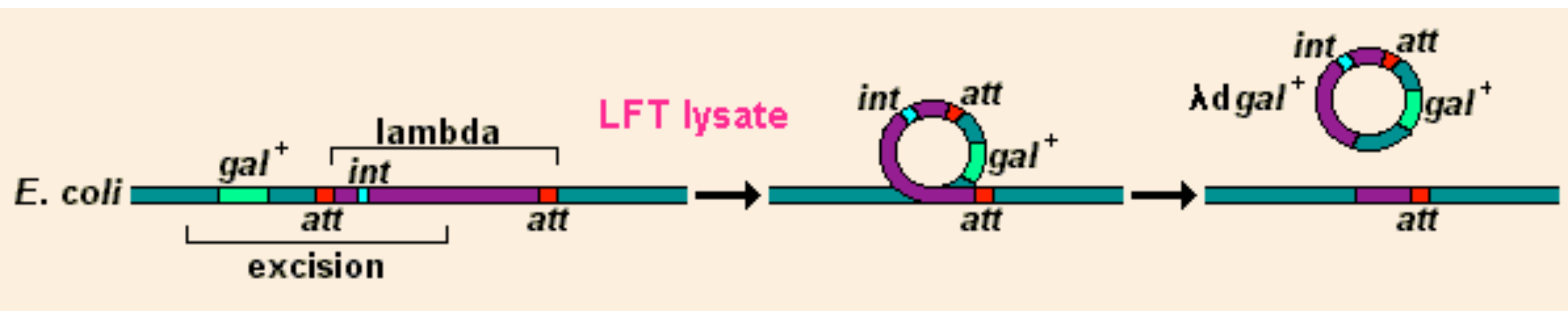
- The recombination must occur between two sites that are the same distance apart as the length the of the phage genome



High-Frequency Transducing (HFT) Lysate: produces phage that can transduce bacterial genes at a very high frequency

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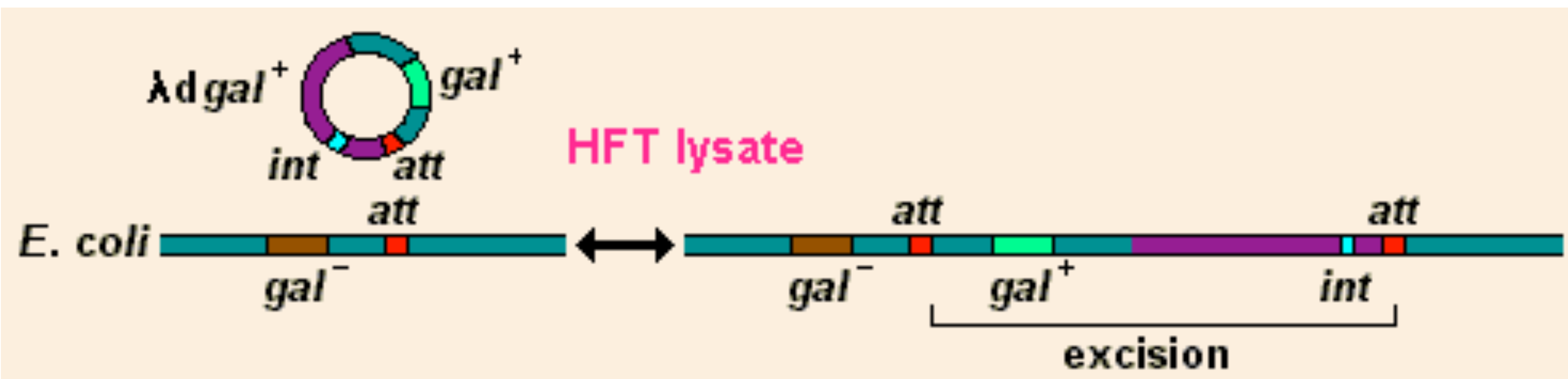
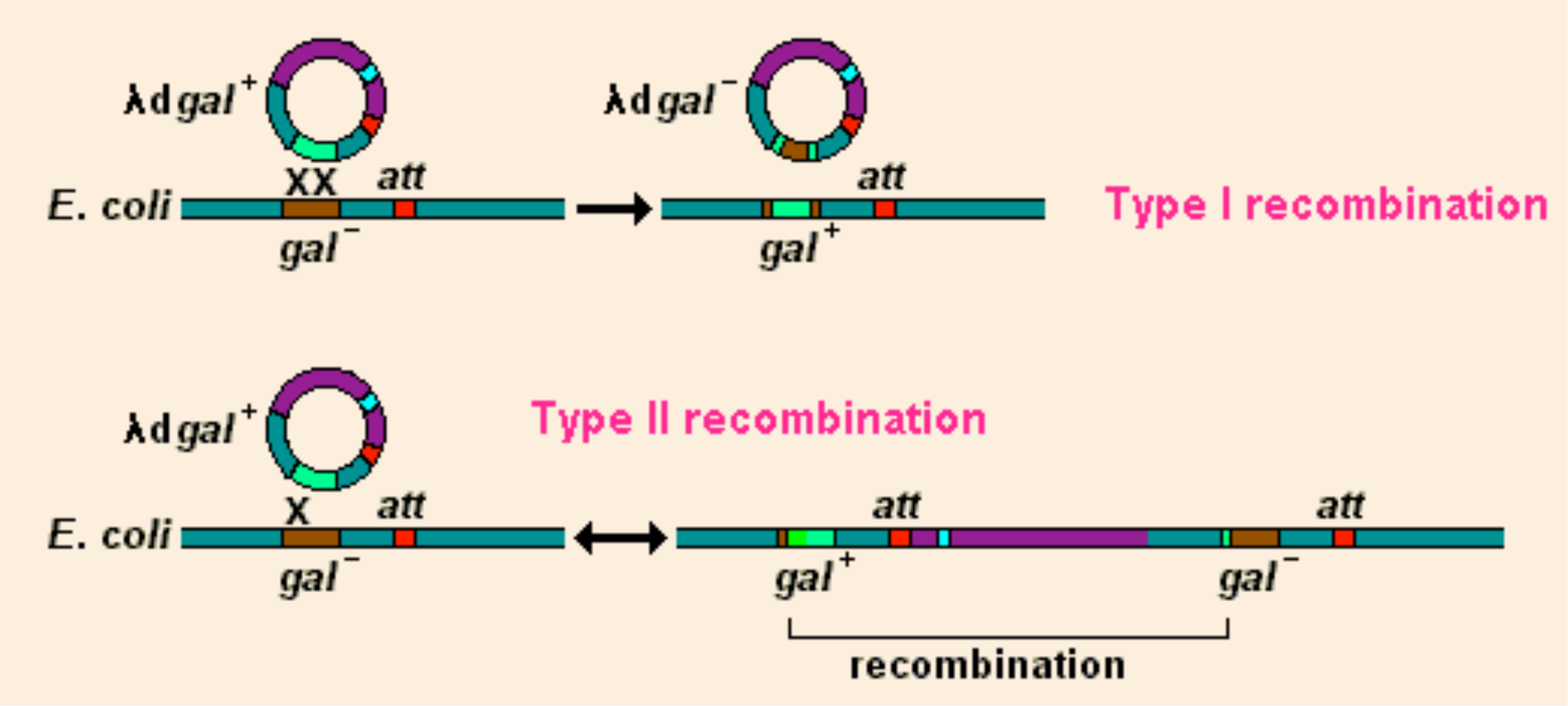
- All phage produced during induction will contain bacterial DNA since the prophage contained bacterial genes (*bio* or *gal*)

- Would only be able to tell if λd*gal* or a λd*bio* transducing particle if host cell is Gal^- or Bio^-



Besides integration of HFT particle into *attB* site, can also have recombination between bacterial DNA



B) Generalized Transduction

1) Transducing particles only contain bacterial DNA

2)

3) Amount of DNA packaged is dependent on phage type

4) For recipient to retain DNA from transducing particle, recombination must occur

Fig. 7.33

Generalized transduction can be used to map the location of chromosomal genes

Frequency of cotransduction is inversely proportional to the distance between 2 genes

- **Genes that are close together will be cotransduced more often**

  ↳

- **Genes that are far apart on the host chromosome will take more cross-over events for transfer**

  ↳

Example P1 Mapping Problem

Donor: *proT$^+$ metE$^+$ bioP$^+$*

Recipient*: proT$^-$ metE$^-$ bioP$^-$*

Resulting transductants

proT	*metE*	*bioP*	# colonies
+	+	+	50
+	+	-	75
+	-	+	1
+	-	-	300

Cotransducing frequencies:

***proT/metE*:**

***proT/bioP*:**

Cotransducing frequencies can be used to calculate distance between two markers

$d = L(1 - CF^{1/3})$

d: distance between markers

L:

CF: cotransducing frequency

Cotransducing frequencies:

***proT/metE*: = 0.29**

***proT/bioP*: = 0.12**

***proT/metE* distance =**

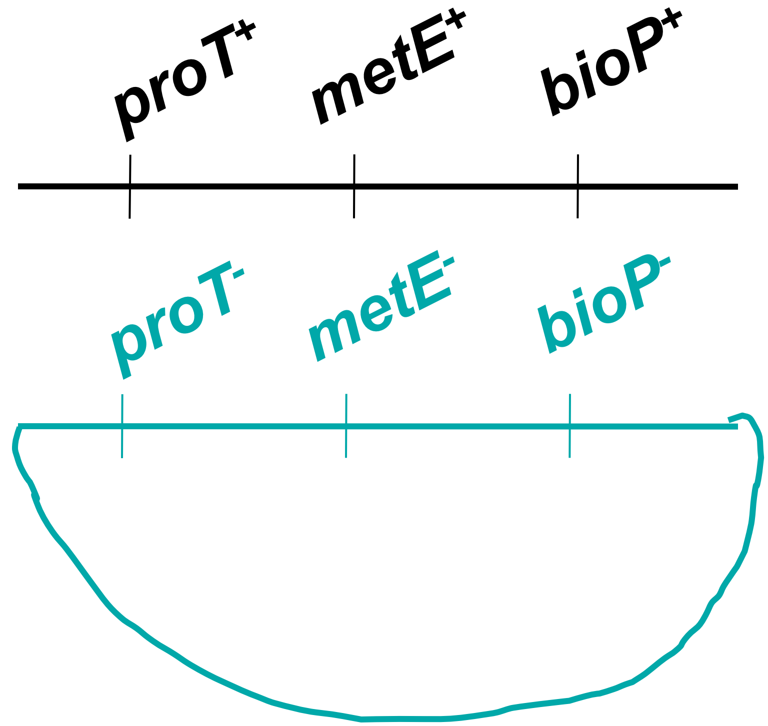
***proT/bioP* distance =**

Determining the gene order: look for recipient marker in the least frequent category (will be in middle)

Donor: ***proT*⁺ *metE*⁺ *bioP*⁺**

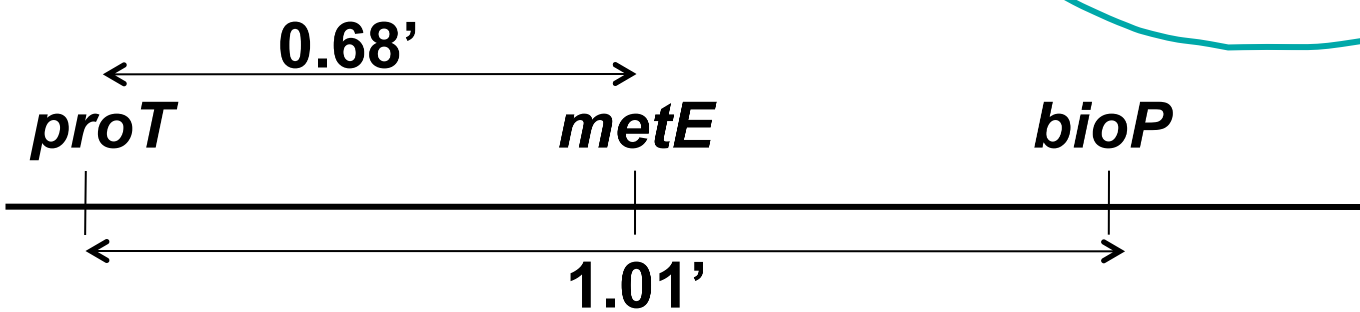
Recipient: *proT*⁻ *metE*⁻ *bioP*⁻

proT	*metE*	*bioP*	# colonies
+	+	+	50
+	+	-	75
+	-	+	1
+	-	-	300



***proT/metE* distance =**

***proT/bioP* distance =**



P1 phage is used in a generalized transduction experiment with the following *E. coli* strains:

Donor: *aroF*- *hisA*+ *thrB*-
Recipient: *aroF*+ *hisA*- *thrB*+

aroF	***hisA***	***thrB***	**#of colonies**
-	+	+	330
-	+	-	127
+	+	-	41
+	+	+	514

A) What are the cotransducing frequencies between the selected marker and both unselected markers?

Donor: ***aroF- hisA+ thrB-***
Recip: ***aroF+ hisA- thrB+***

aroF	*hisA*	*thrB*	#of colonies
-	+	+	330
-	+	-	127
+	+	-	41
+	+	+	514

B) What are the corresponding distances between the selected marker and the unselected markers?

C) Draw a map of the *E.coli* chromosome in this region, using the proper units.