2 factor & 3 factor Crosses

Given: 3 markers, m_1, m_2 & m_3

3 2 factor crosses

$m_1 \times m_2 = rf_{1,2}$ e.g. $= 0.1$

$m_1 \times m_3 = rf_{1,3} = 0.26$

$m_2 \times m_3 = rf_{2,3} = 0.2$

If $rf_{1,3} > rf_{2,3} > rf_{1,2}$

Order = 1 — 2 — 3

N.B. rf's not additive

$rf_{1,3}(0.26) \neq rf_{1,2}(0.1) + rf_{2,3}(0.2)$

$rf_{1,3} = rf_{1,2}(1 - rf_{2,3}) + rf_{2,3}(1 - rf_{1,2})$

$= rf_{1,2} + rf_{2,3} - 2\, rf_{1,2} rf_{2,3}$

$= .1 + .2 - 2(.1 \times .2) = .26$

Establish map with 1 3 factor cross

	$m_1\ m_2\ m_3^+$	} parentals 72%	
	$m_1^+\ m_2^+\ m_3$		
switch of 1st factor	$m_1^+\ m_2\ m_3^+$	8%	} Recomb.
	$m_1\ m_2^+\ m_3$		
switch of 2nd factor	$m_1\ m_2^+\ m_3^+$	2%	
	$m_1^+\ m_2\ m_3$		
switch of 3rd factor	$m_1\ m_2\ m_3$	18%	
	$m_1^+\ m_2^+\ m_3^+$		

$rf_{12} = 8\% + 2\% = 10\%$

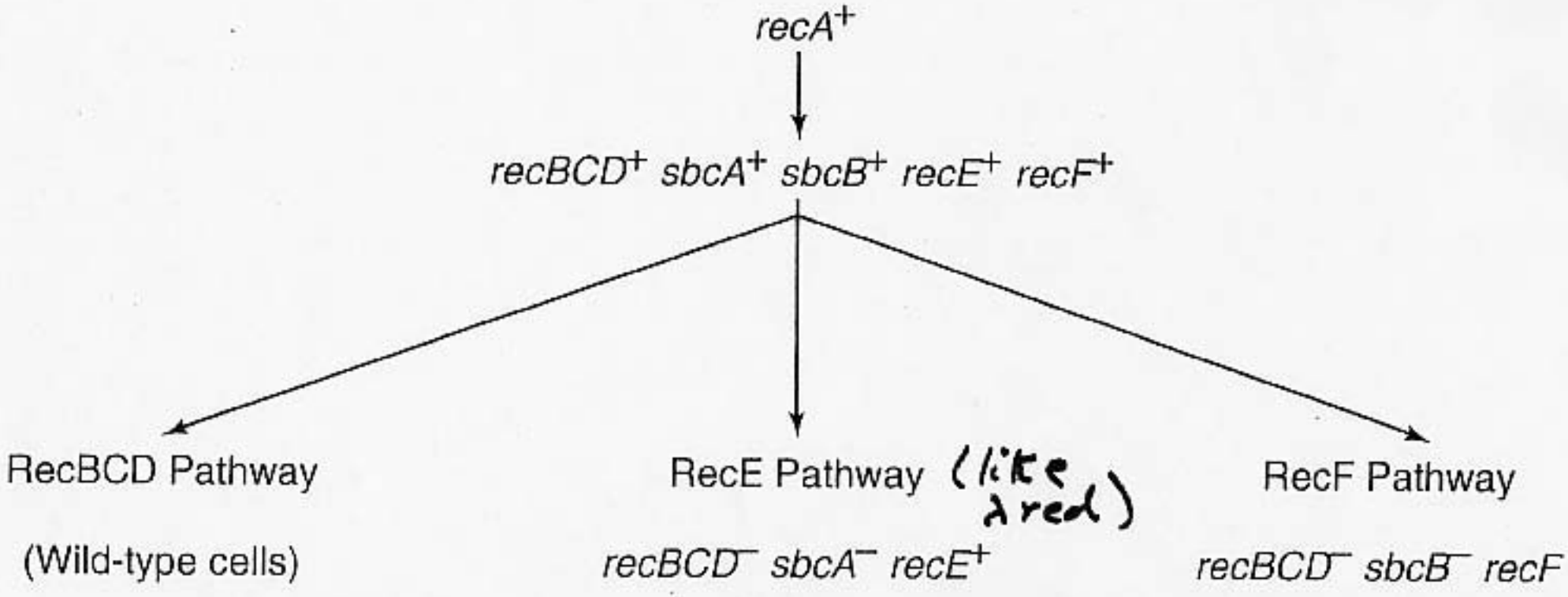
$rf_{23} = 18\% + 2\% = 20\%$

$rf_{13} = 18\% + 8\% = 26\%$

Table 10.2

TABLE 10.2 Analogy between phage and host recombination functions

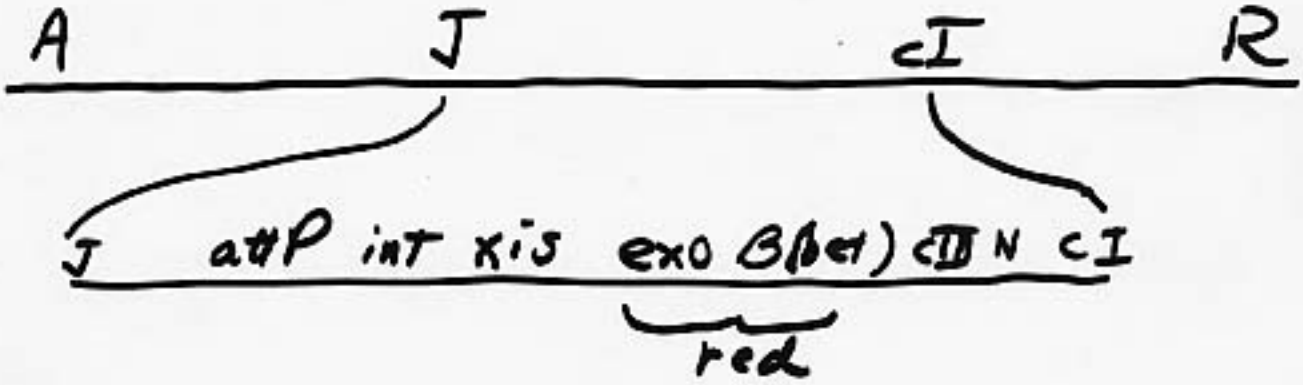
Phage function	Analogous *E. coli* function
T4 UvsX	RecA
T4 gene 49	RuvC
T7 gene 3	RuvC
T4 genes 46 and 47	RecBCD
λ ORF in *nin* region	RecO, RecR, RecF
Rac *recE* gene	RecJ, RecQ
λ *gam*	Inhibits RecBCD
λ *exo*	RecBCD, RecJ
λ *bet*	RecA
rusA (DLP12 prophage)	RuvC



sbc = suppressor recBC

sbcA + recE on defective prophage Rac
RecE = exonuclease VIII

λ Recombination



Red - general
int, xis - site-specific

Red:

λ A_{am} red+ × λ J_{am} red+ or
λ A_{am} red⁻ × λ J_{am} red⁻

λ	Host		
	Rec^+	$RecA^-$	$RecB^-$
red+	.8	1.1	0.7
red⁻	.2	.006	.02

Int :

$$\frac{Jam \quad (attP)\ cI^{+} \qquad R^{+}}{\times}$$

$$J^{+} \qquad (attP)\ cI \qquad Ram$$

Select on E. coli Su^{-}

$\therefore$ $J^{+}cI^{+}R^{+}$ (x between J + cI)

J^{+} cI R^{+} (x between cI + R)

Phage		Host	% Recomb	
red	int	rec	JcI	cI R
+	+	+	7.5	3.6
+	+	−	7.8	3.1
+	−	−	4.1	3.0
−	+	−	2.0	< 0.05
−	−	−	≤ .05	≤ .05

Figure 10.9

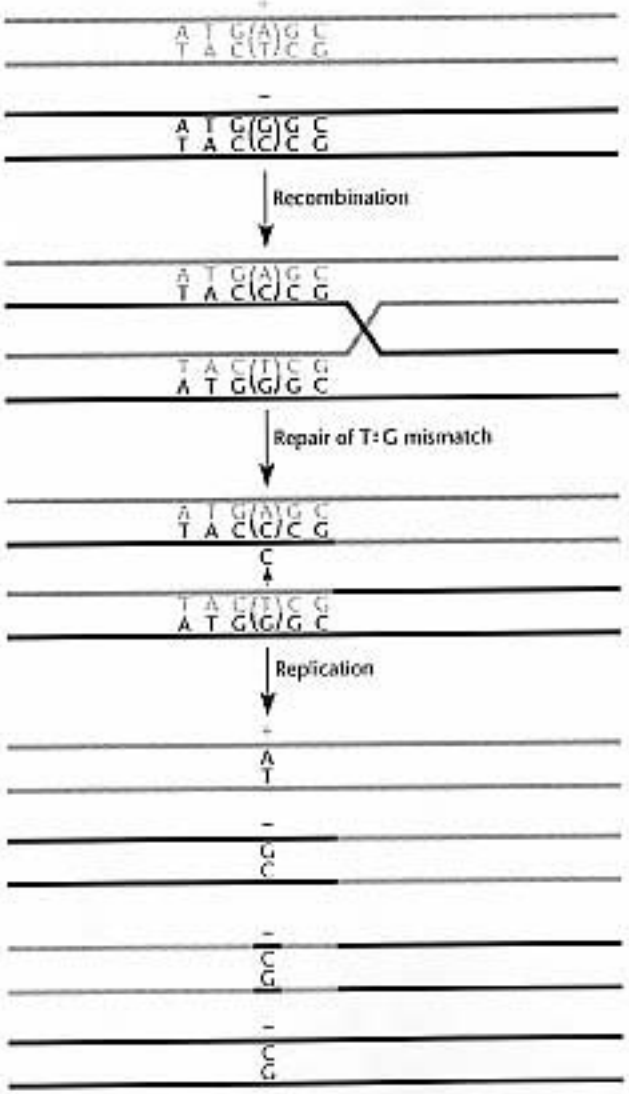
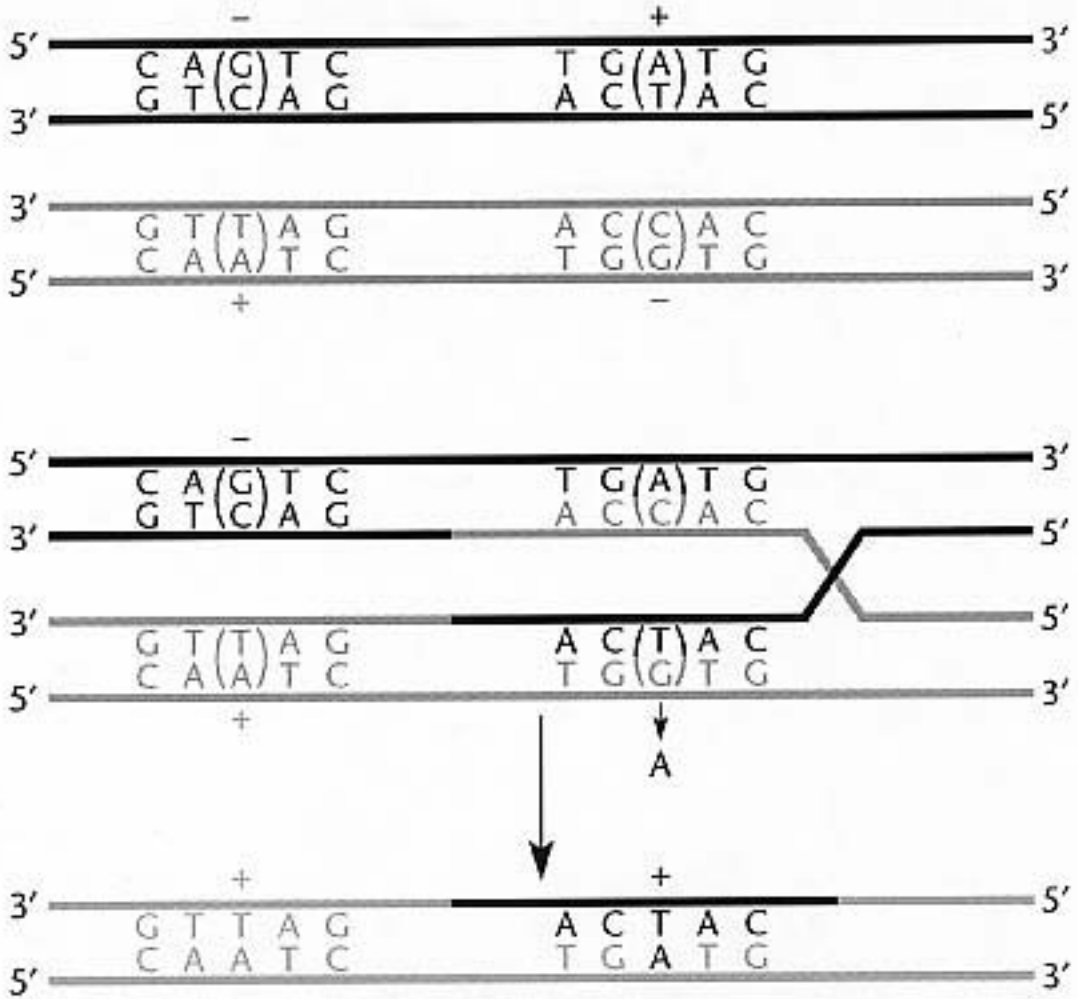
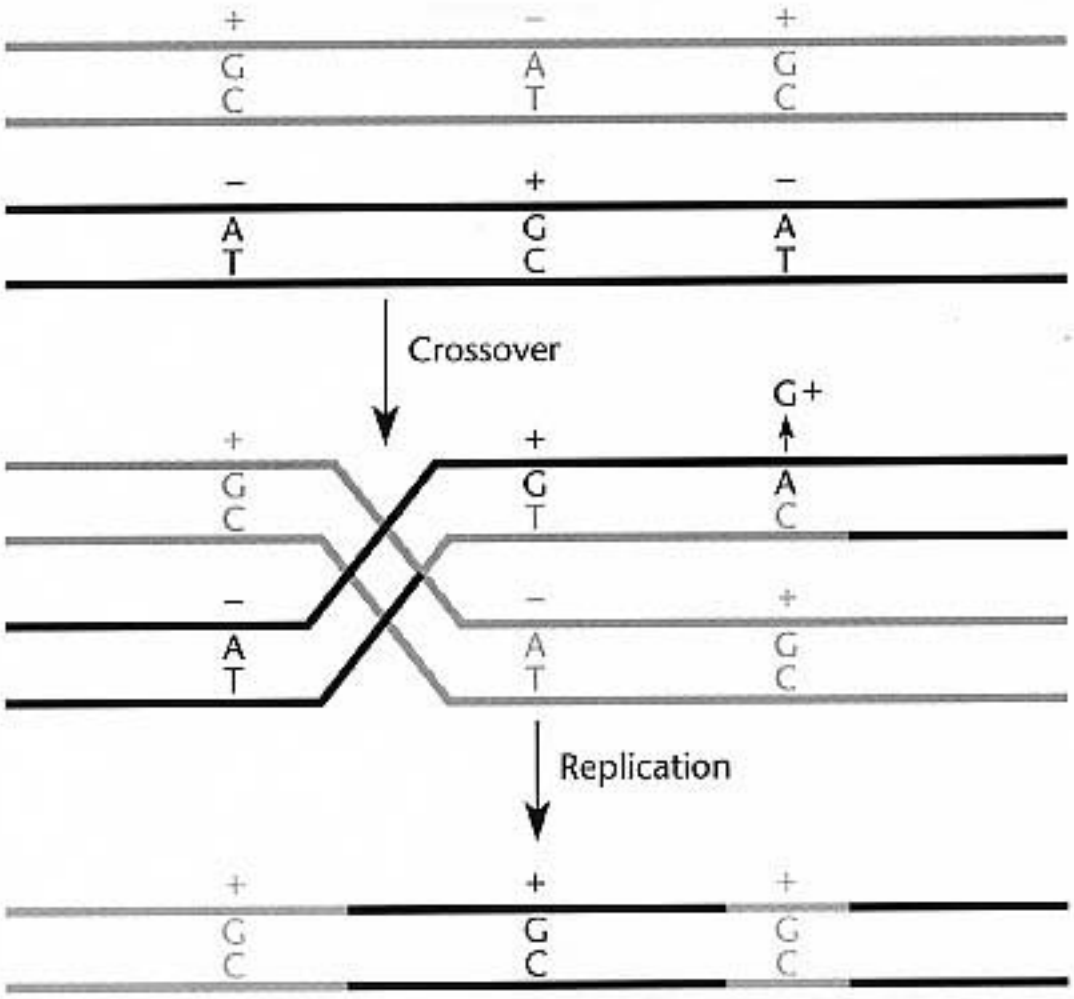


Figure 10.10



Wild-type recombinant

Figure 10.11


+
G
C
–
A
T
+
G
C
–
A
T
+
G
C
–
A
T
Crossover
G+
+
G
T
–
A
T
+
A
C
+
G
C
Replication
+
G
C
+
G
C
+
G
C
Wild-type recombinant