Figure 3.1

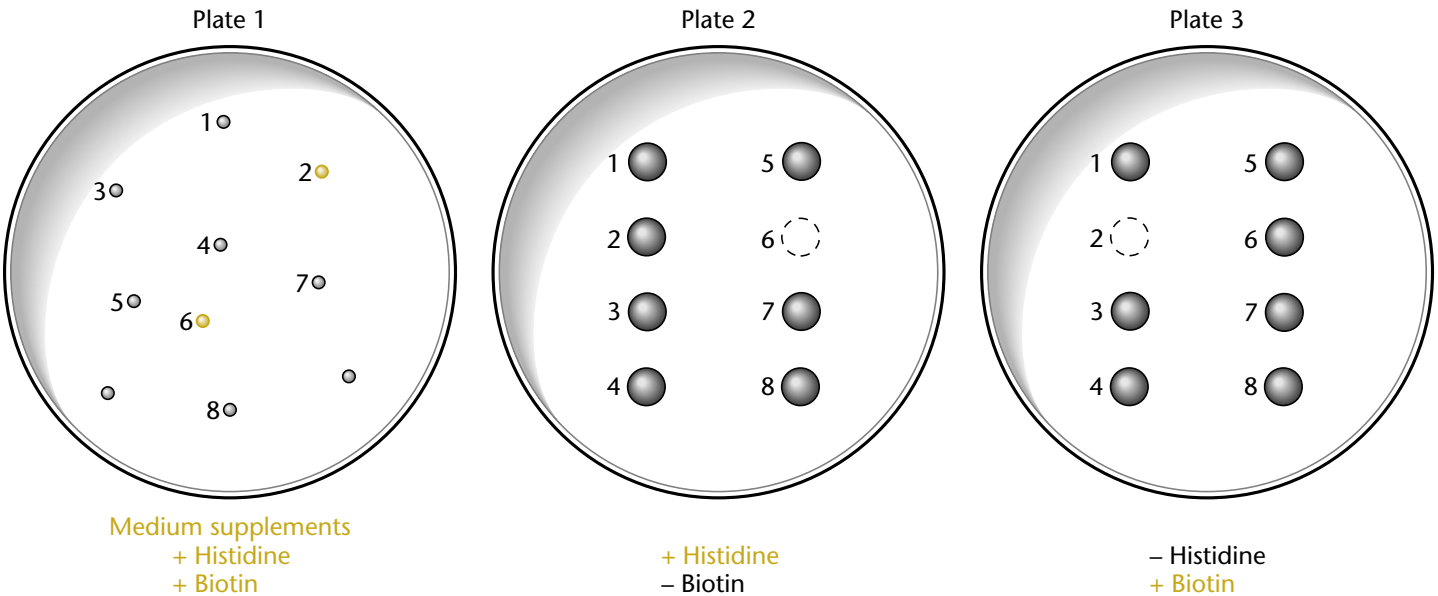


Plate 1
1
2
3
4
5
6
7
8
Medium supplements
+ Histidine
+ Biotin
Plate 2
1
2
3
4
5
6
7
8
+ Histidine
– Biotin
Plate 3
1
2
3
4
5
6
7
8
– Histidine
+ Biotin

Table 3.1

TABLE 3.1 Some resistance mutations

Substance	Toxicity	Resistance mutation
Bacteriophage T1	Infects and kills	Inactivates *tonB* outer membrane protein; phage cannot absorb
Streptomycin	Binds to ribosomes; inhibits translation	Changes ribosomal protein S12 so that it no longer binds
Chlorate	Converted to chlorite, which is toxic	Inactivates nitrate reductase, which converts chlorate to chlorite
High concentrations of valine, no isoleucine	Feedback inhibits acetolactate synthetase; starves for isoleucine	Activates a valine-insensitive acetolactate synthetase

Figure 3.2

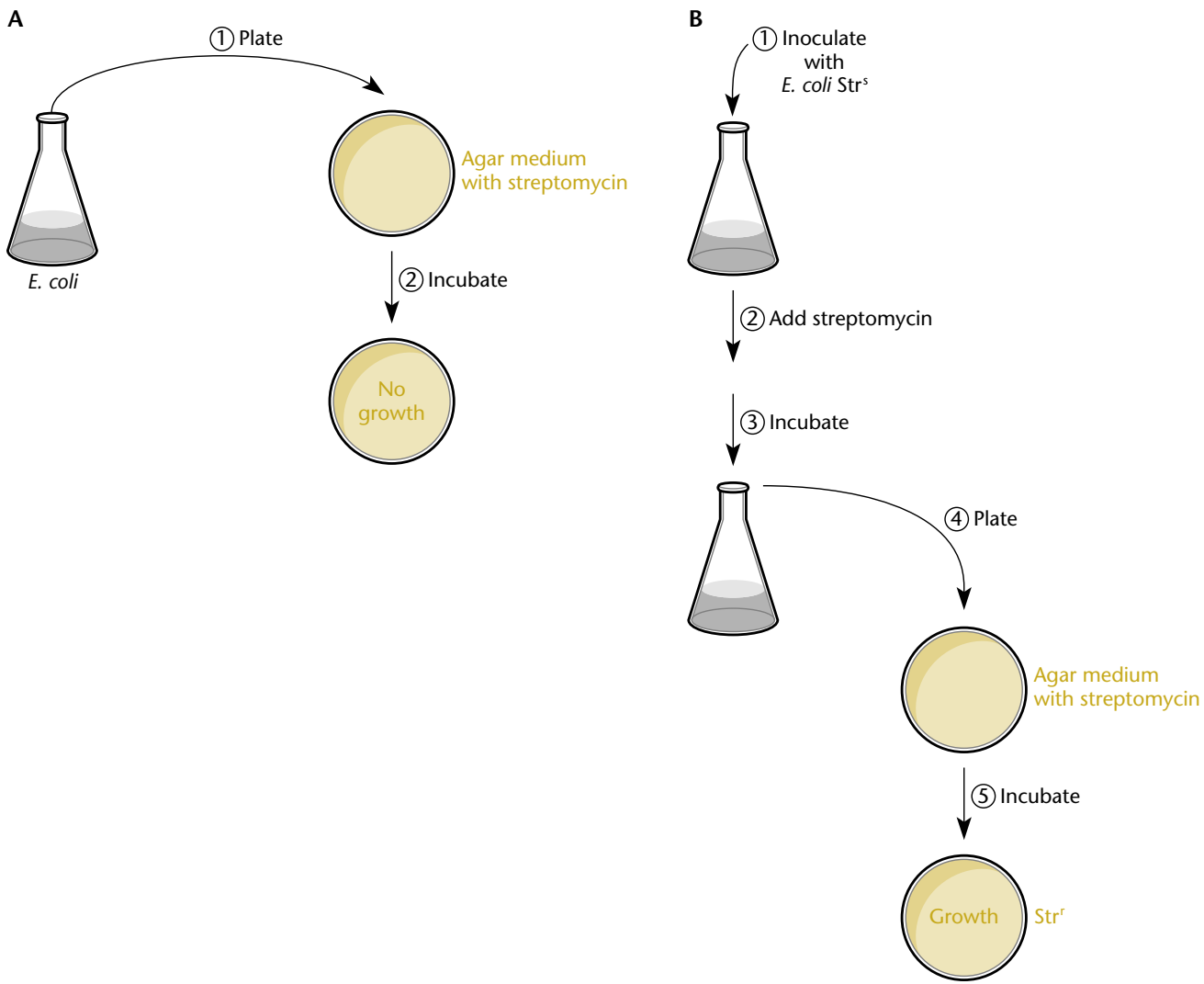

A
1 Plate
E. coli
Agar medium
with streptomycin
2 Incubate
No
growth
B
1 Inoculate
with
E. coli Str^s
2 Add streptomycin
3 Incubate
4 Plate
Agar medium
with streptomycin
5 Incubate
Growth
Str^r

Figure 3.3

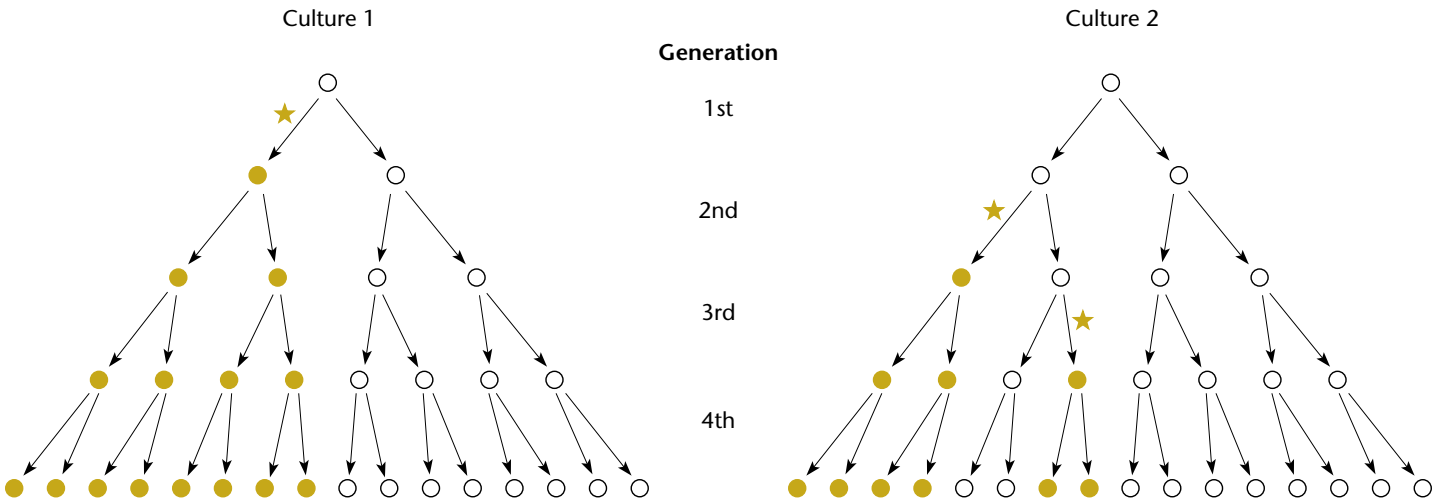


Figure 3.4

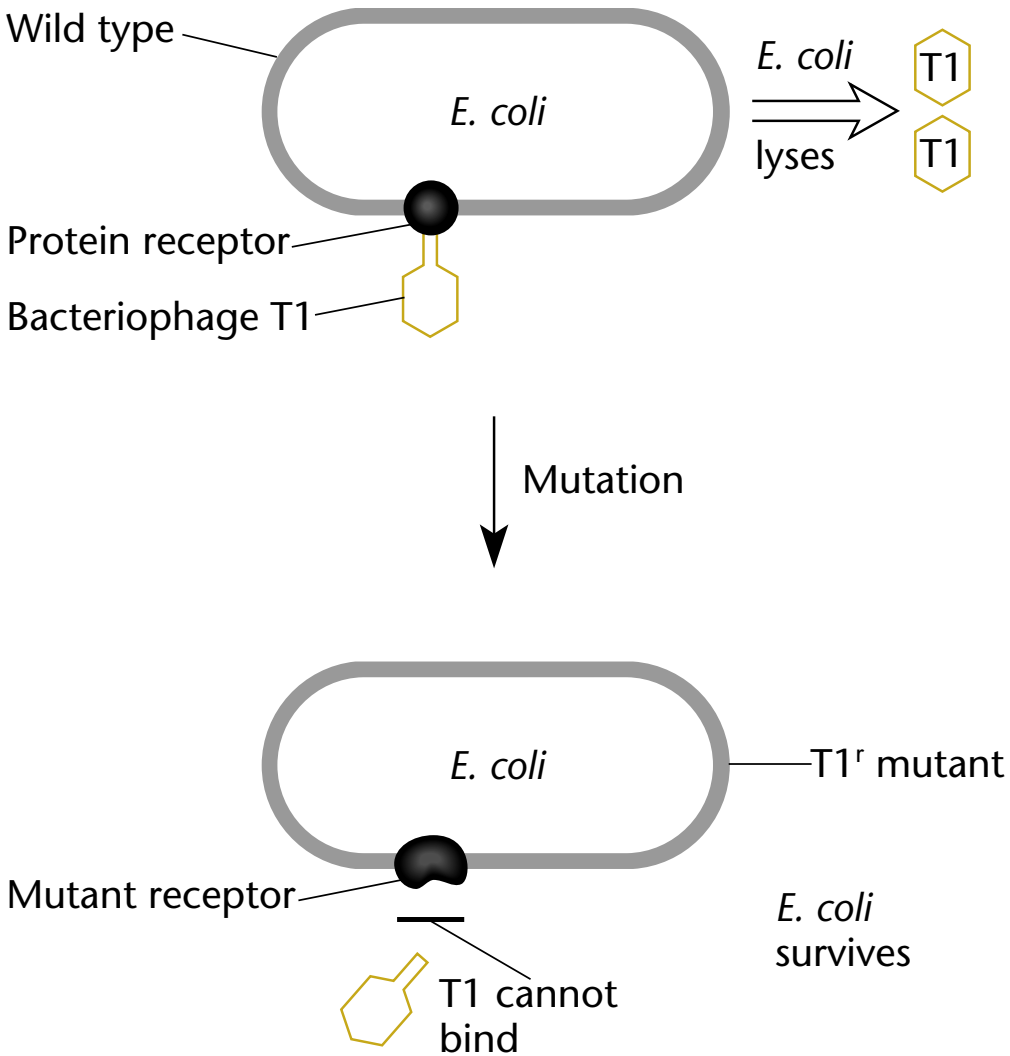

Wild type
E. coli
E. coli
lyses
T1
T1
Protein receptor
Bacteriophage T1
Mutation
E. coli
T1r mutant
Mutant receptor
E. coli
survives
T1 cannot
bind

Figure 3.5

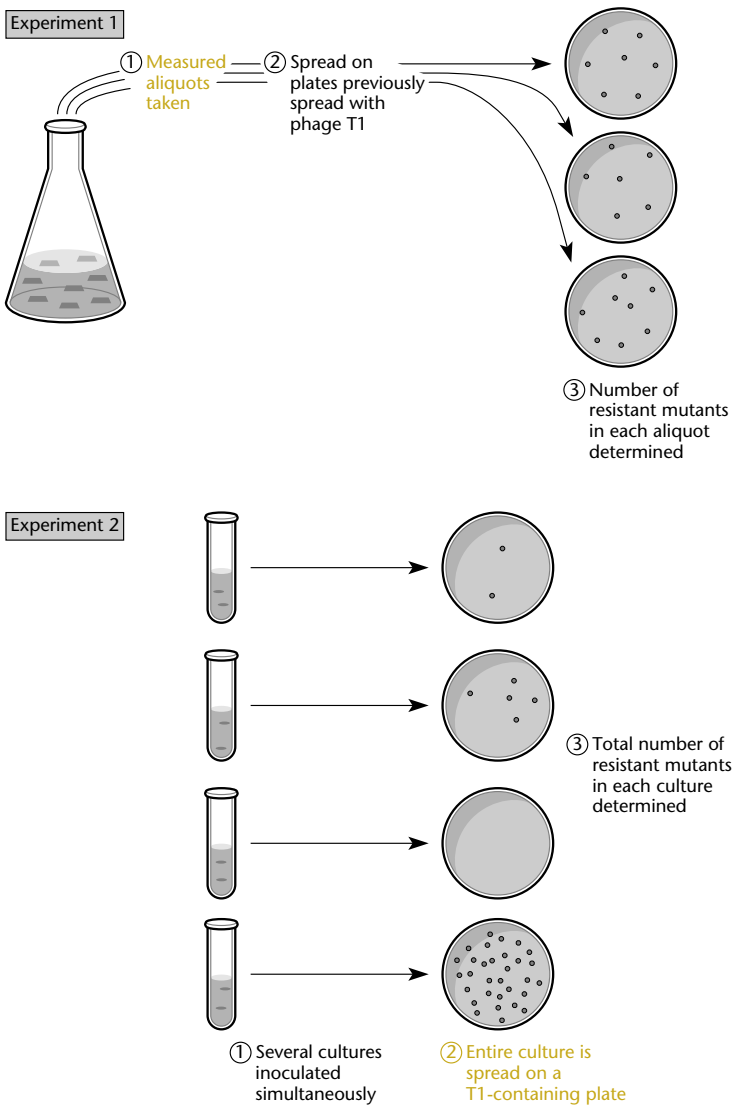

Experiment 1
① Measured aliquots taken
② Spread on plates previously spread with phage T1
③ Number of resistant mutants in each aliquot determined
Experiment 2
③ Total number of resistant mutants in each culture determined
① Several cultures inoculated simultaneously
② Entire culture is spread on a T1-containing plate

Table 3.2

TABLE 3.2 The Luria and Delbrück experiment

Experiment 1		Experiment 2	
Aliquot no.	No. of resistant bacteria	Culture no.	No. of resistant bacteria
1	14	1	1
2	15	2	0
3	13	3	3
4	21	4	0
5	15	5	0
6	14	6	5
7	26	7	0
8	16	8	5
9	20	9	0
10	13	10	6
		11	107
		12	0
		13	0
		14	0
		15	1
		16	0
		17	0
		18	64
		19	0
		20	35

Figure 3.6

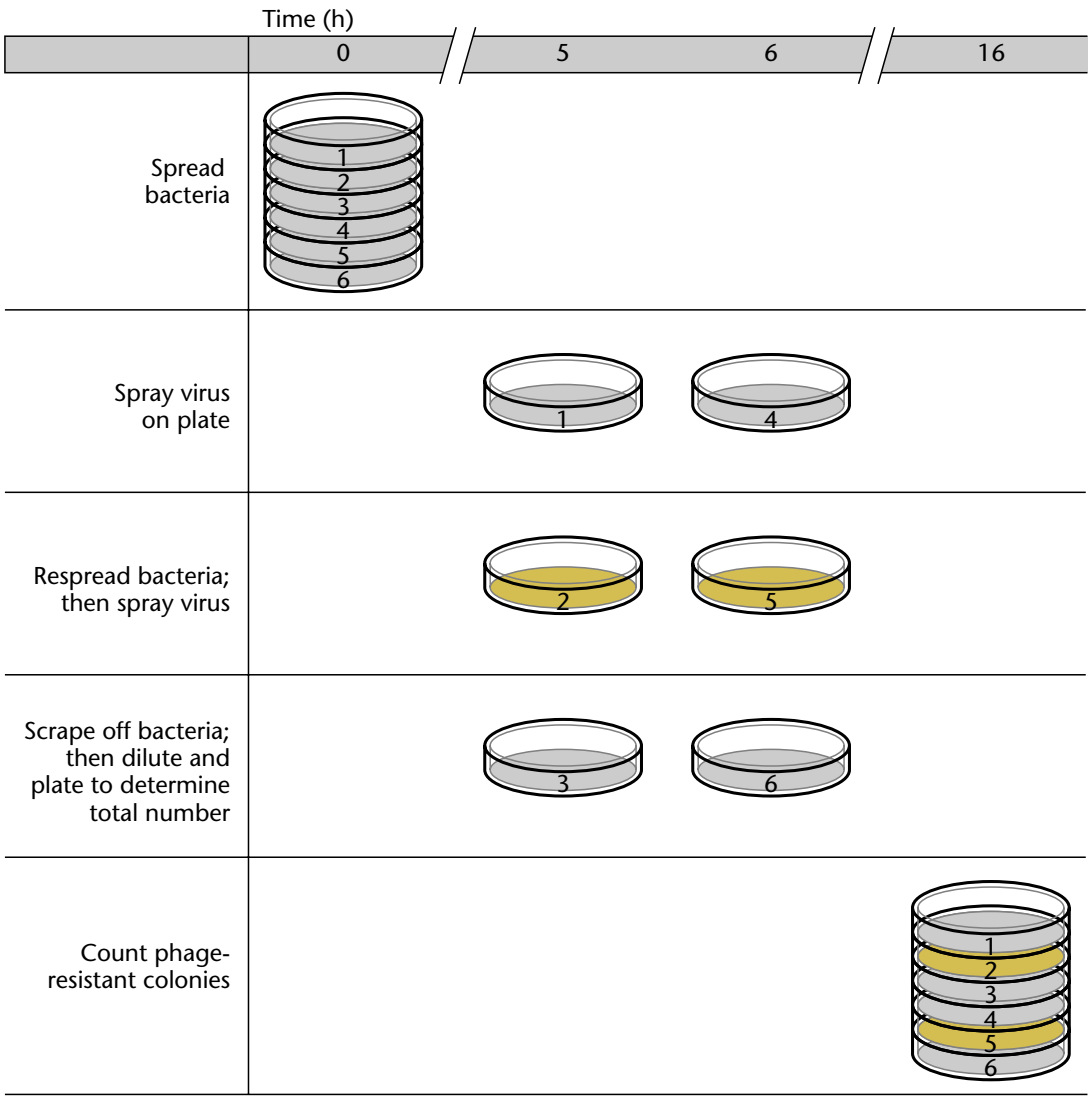

Time (h)
0
5
6
16
Spread bacteria
1
2
3
4
5
6
Spray virus on plate
1
4
Respread bacteria; then spray virus
2
5
Scrape off bacteria; then dilute and plate to determine total number
3
6
Count phage-resistant colonies
1
2
3
4
5
6

Table 3.3

TABLE 3.3 The Newcombe experiment

Incubation time (h)	No. of bacteria plated	Ending no. of bacteria	No. of resistant colonies[a]	
			unsp	sp
5	5.1×10^4	2.6×10^8 (plate 3)	8 (plate 1)	13 (plate 2)
6	5.1×10^4	2.8×10^9 (plate 6)	49 (plate 4)	3,719 (plate 5)

[a]unsp, unspread; sp, spread.

Figure 3.7

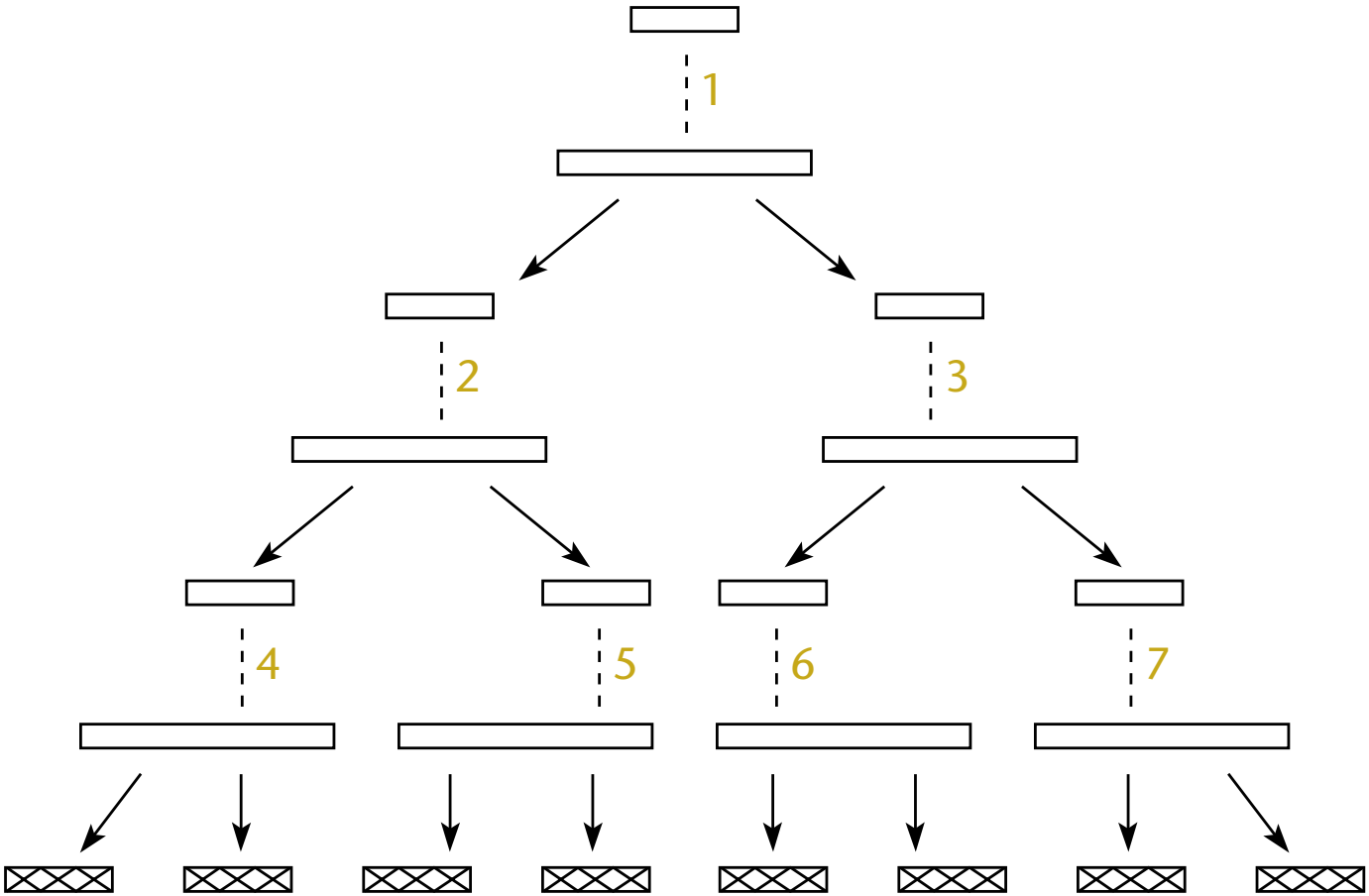


Figure 3.8

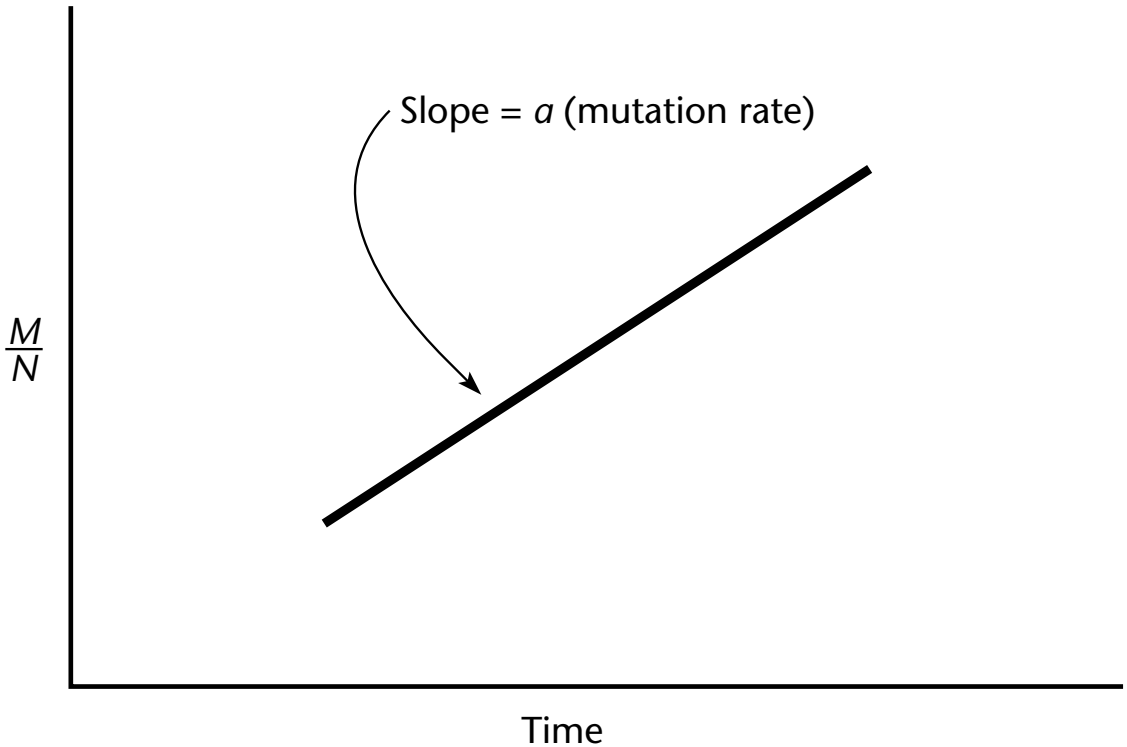


Figure 3.9

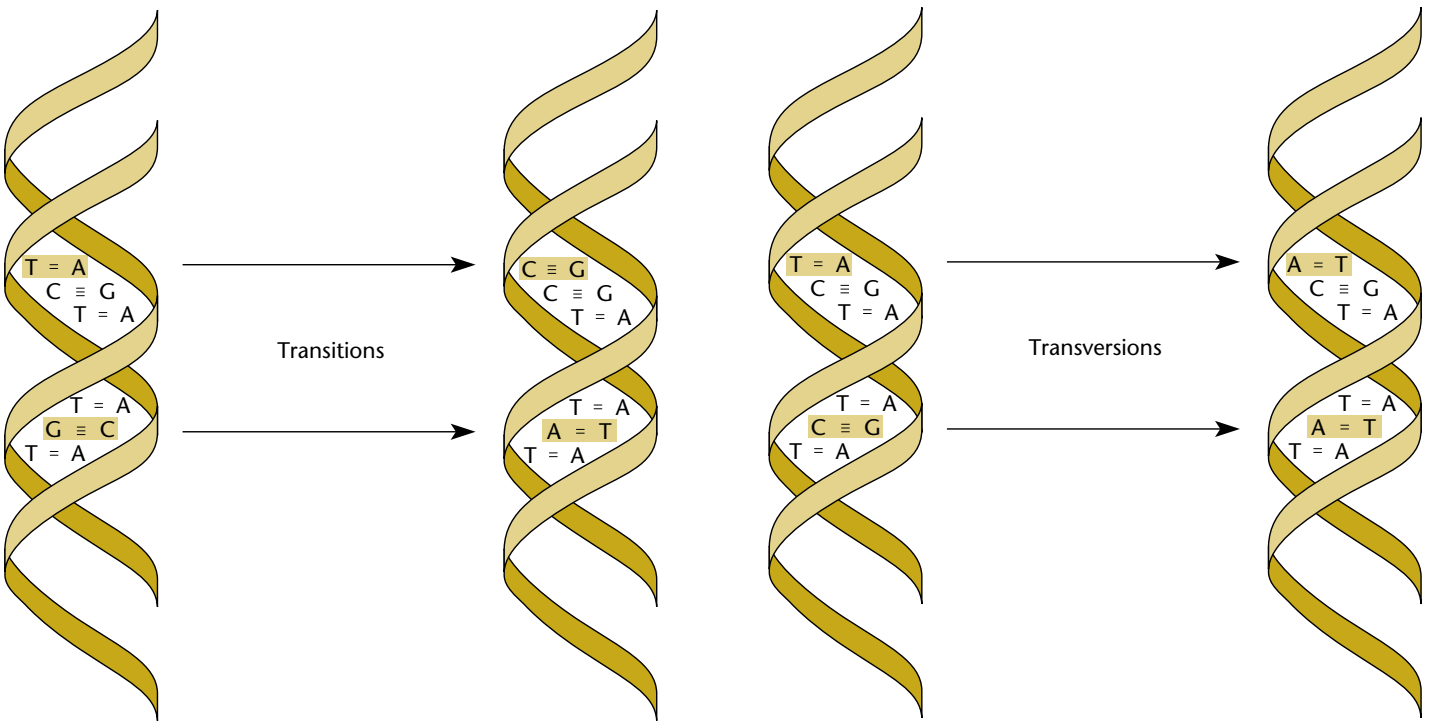

T = A
C ≡ G
T = A
T = A
G ≡ C
T = A
Transitions
C ≡ G
C ≡ G
T = A
T = A
A = T
T = A
T = A
C ≡ G
T = A
T = A
C ≡ G
T = A
Transversions
A = T
C ≡ G
T = A
T = A
A = T
T = A

Figure 3.10

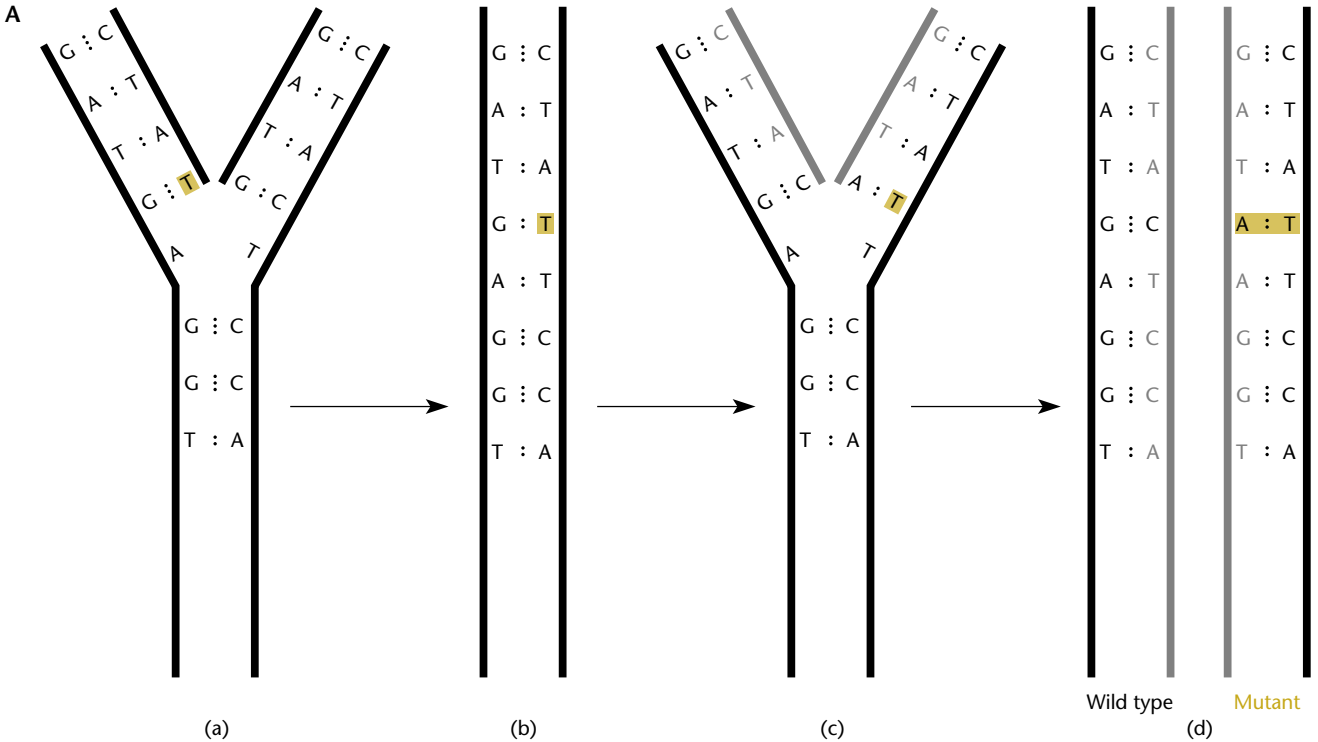

A
(a)
(b)
(c)
Wild type
Mutant
(d)
B
Deoxyribose
Deoxyribose
Guanine
Thymine (enol form)

Figure 3.11

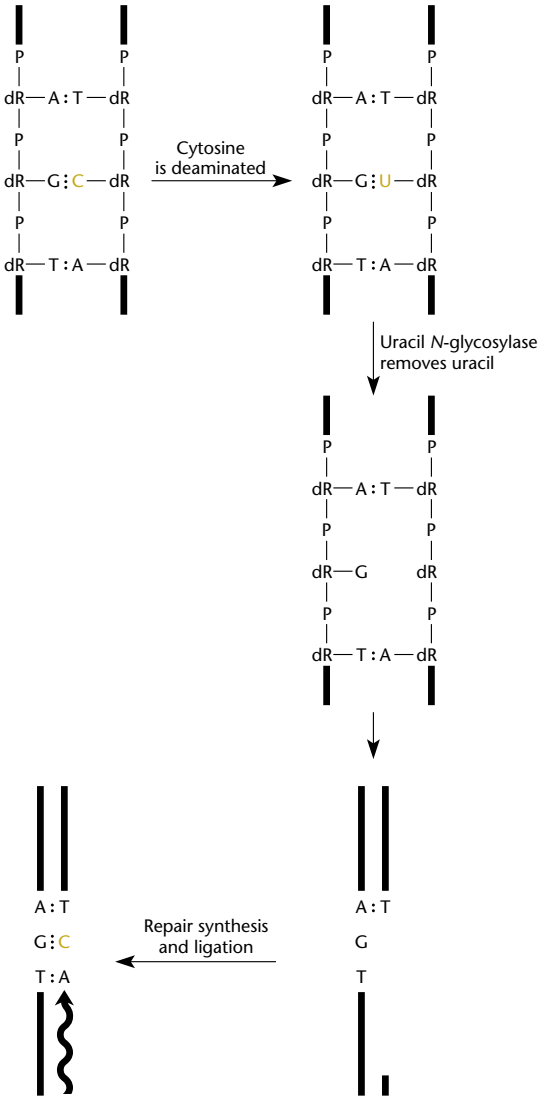

Cytosine
is deaminated
Uracil N-glycosylase
removes uracil
Repair synthesis
and ligation
A:T
G:C
T:A
A:T
G
T

Figure 3.12

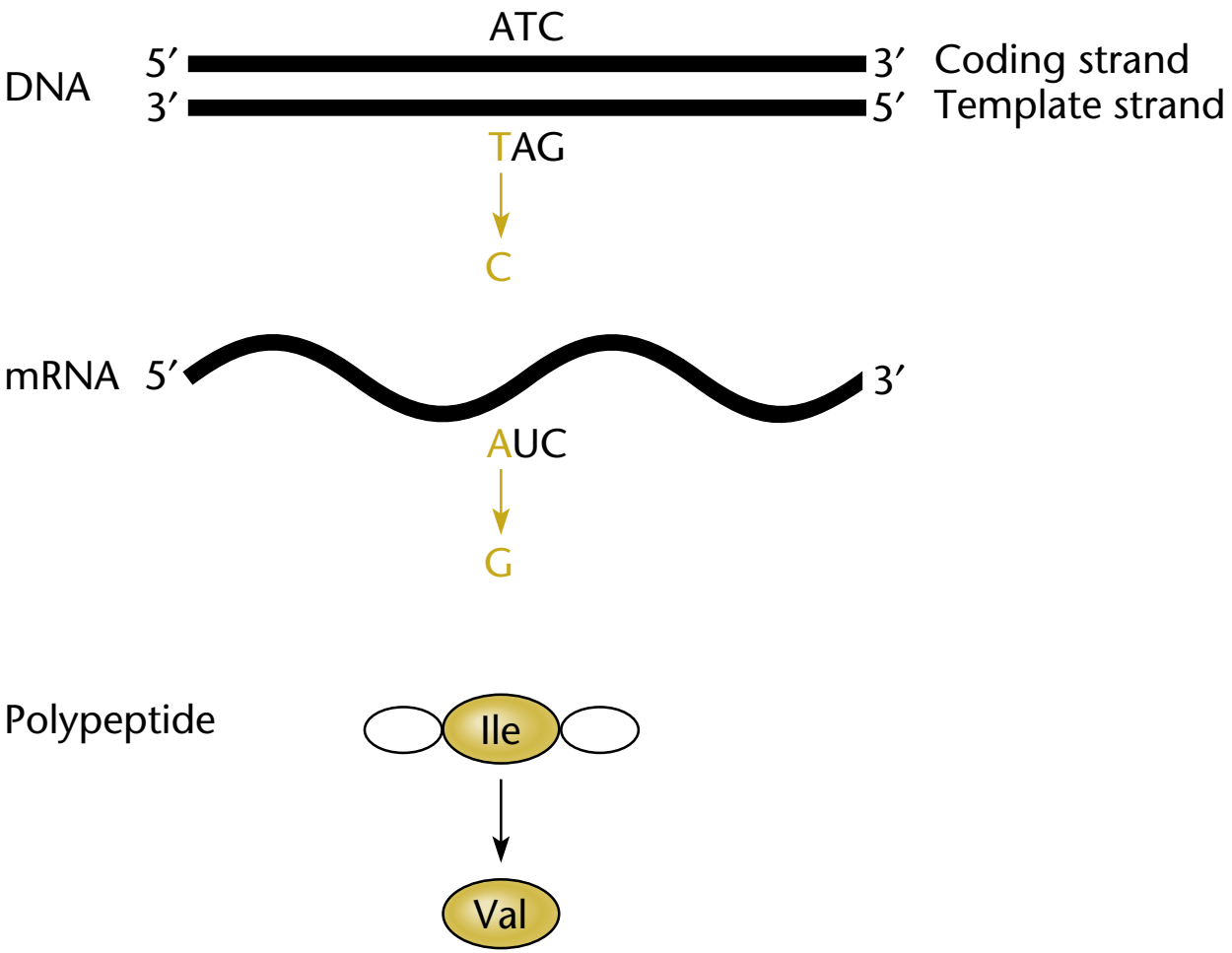

ATC
DNA
5′
3′
3′
5′
Coding strand
Template strand
TAG
C
mRNA
5′
3′
AUC
G
Polypeptide
Ile
Val

Figure 3.13

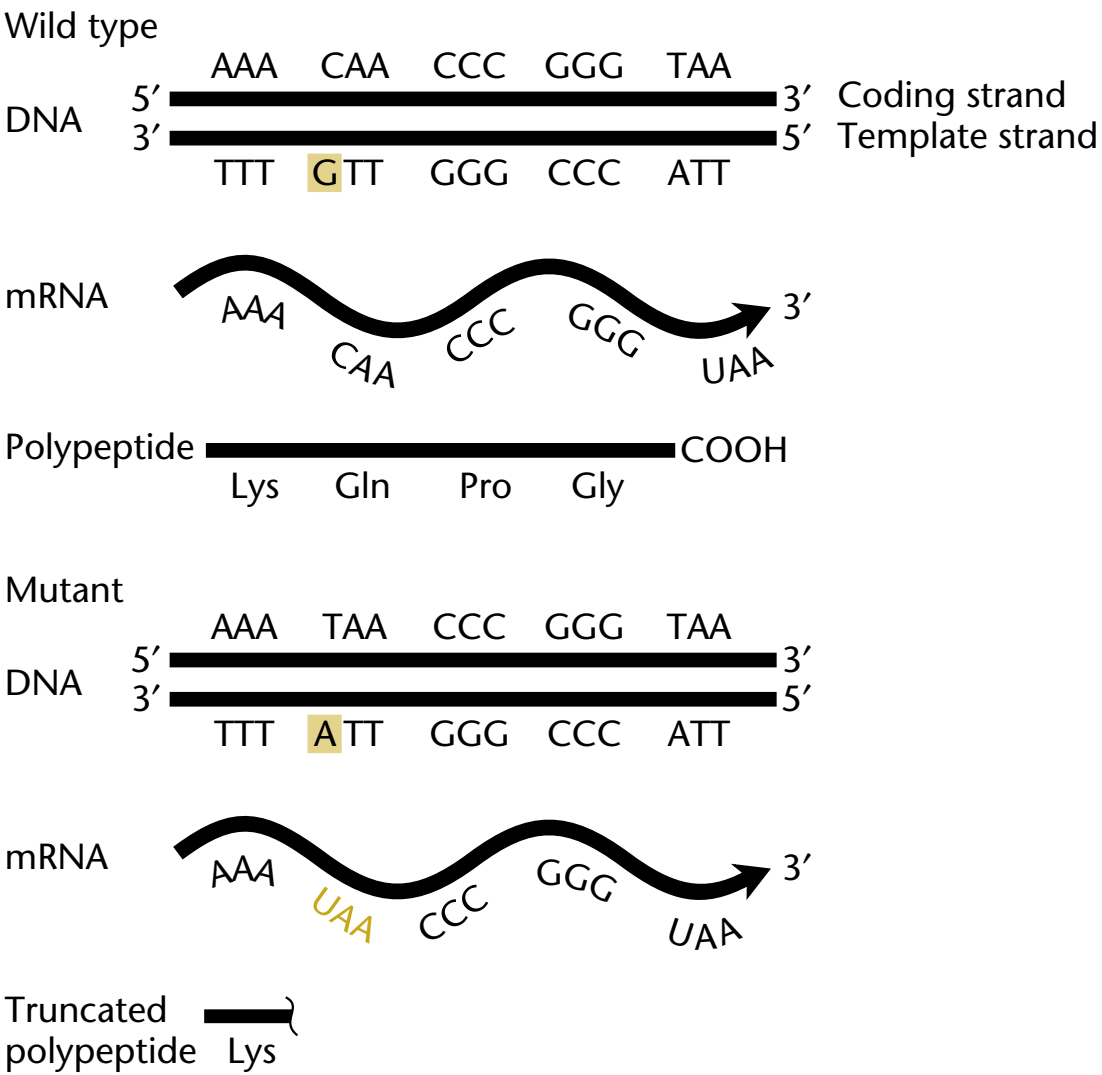

Wild type
AAA CAA CCC GGG TAA
DNA
5′
3′
3′
5′
Coding strand
Template strand
TTT GTT GGG CCC ATT
mRNA
AAA
CAA
CCC
GGG
UAA
3′
Polypeptide
COOH
Lys Gln Pro Gly
Mutant
AAA TAA CCC GGG TAA
DNA
5′
3′
3′
5′
TTT ATT GGG CCC ATT
mRNA
AAA
UAA
CCC
GGG
UAA
3′
Truncated polypeptide
Lys

Figure 3.14

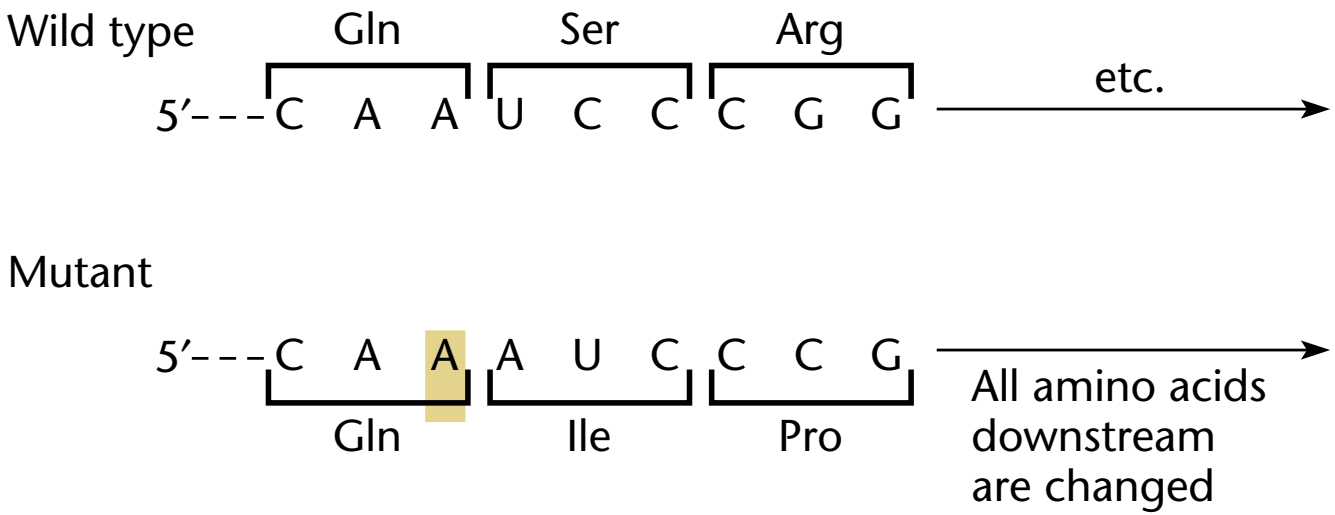

Wild type
Gln
Ser
Arg
5′– – –C A A U C C C G G
etc.
Mutant
5′– – –C A A A U C C C G
Gln
Ile
Pro
All amino acids downstream are changed

Figure 3.15

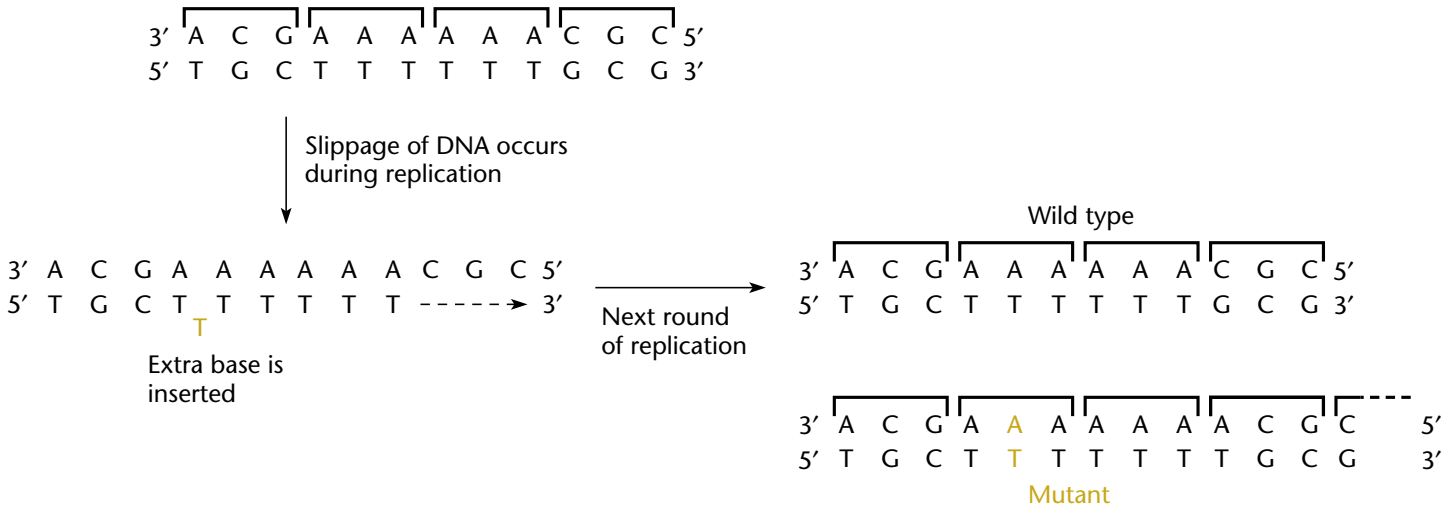

3′ A C G A A A A A A C G C 5′
5′ T G C T T T T T T G C G 3′
Slippage of DNA occurs during replication
3′ A C G A A A A A A C G C 5′
5′ T G C T T T T T T 3′
T
Extra base is inserted
Next round of replication
Wild type
3′ A C G A A A A A A C G C 5′
5′ T G C T T T T T T G C G 3′
3′ A C G A A A A A A C G C 5′
5′ T G C T T T T T T G C G 3′
Mutant

Figure 3.16

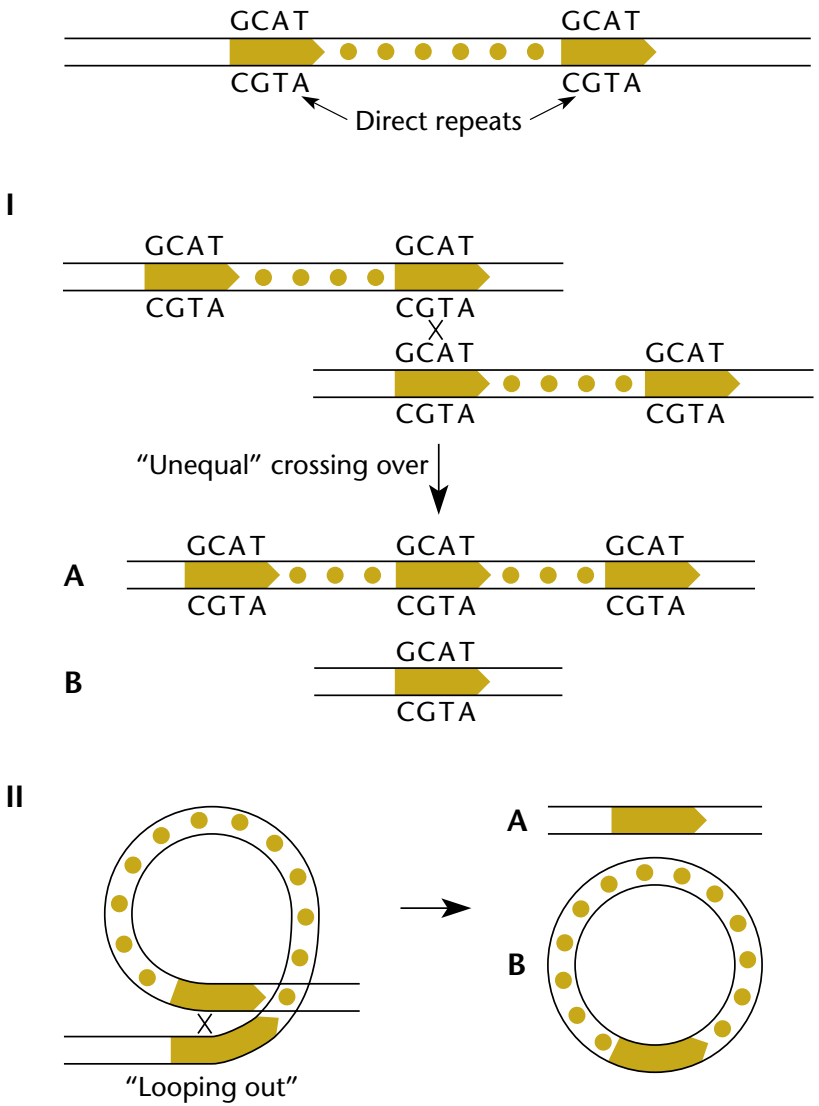
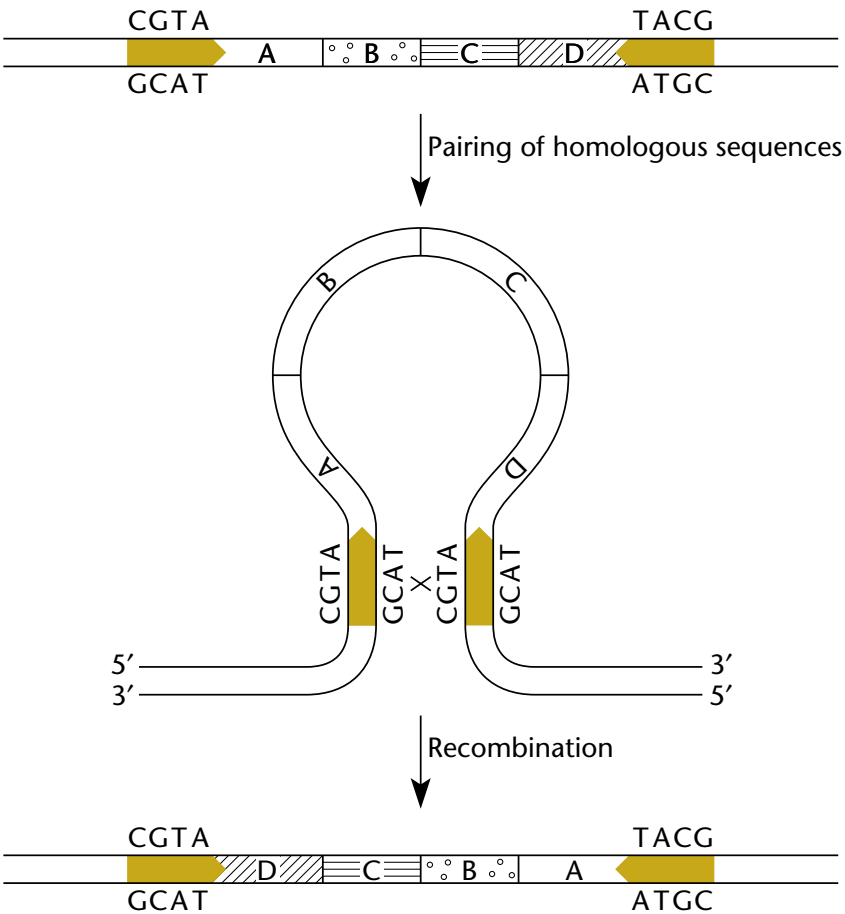

GCAT
GCAT
CGTA
CGTA
Direct repeats
I
GCAT
GCAT
CGTA
CGTA
GCAT
GCAT
CGTA
CGTA
"Unequal" crossing over
A
GCAT
GCAT
GCAT
CGTA
CGTA
CGTA
B
GCAT
CGTA
II
A
B
"Looping out"

Figure 3.17


CGTA
A
B
C
D
TACG
GCAT
ATGC
Pairing of homologous sequences
B
C
A
D
CGTA
GCAT
CGTA
GCAT
5′
3′
3′
5′
Recombination
CGTA
D
C
B
A
TACG
GCAT
ATGC

Box 3.2

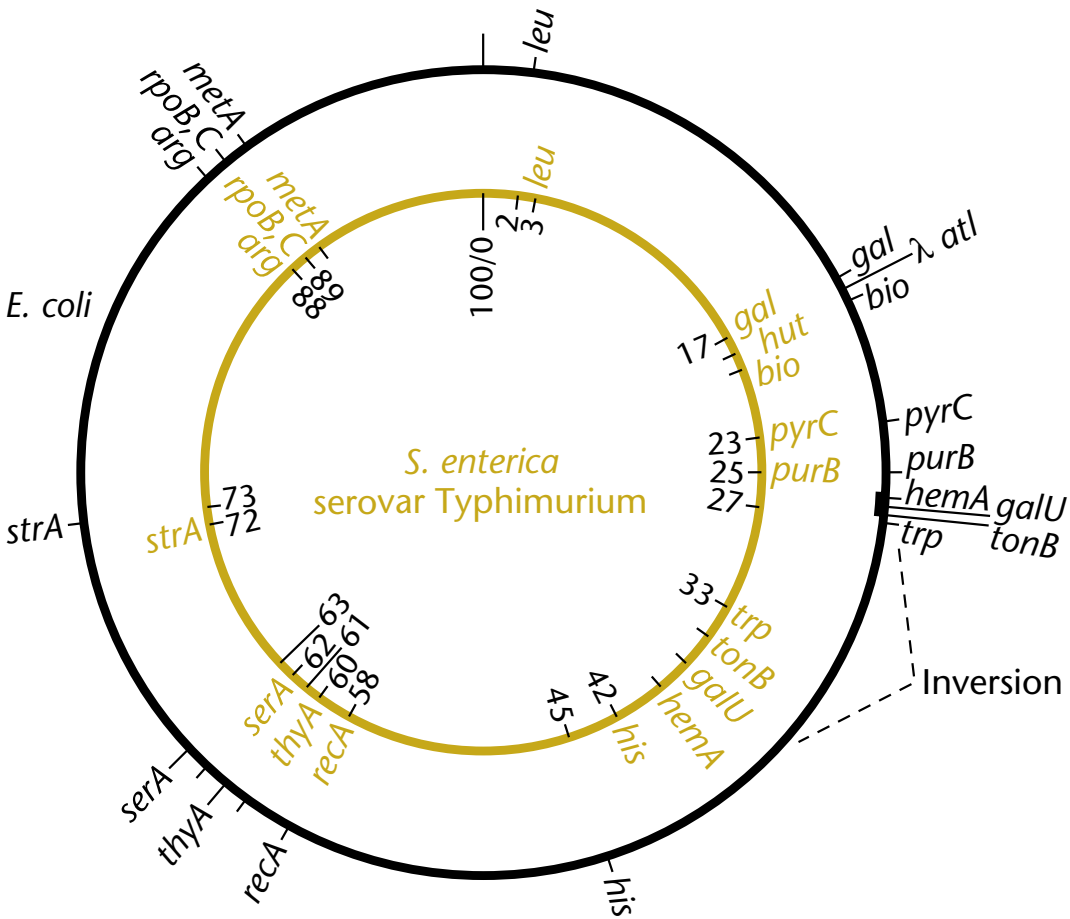

E. coli
S. enterica
serovar Typhimurium
leu
metA
rpoB,C
arg
gal
bio
λ atl
pyrC
purB
hemA
galU
tonB
trp
Inversion
his
recA
thyA
serA
strA
100/0
2
3
88
86
17
hut
23
25
27
33
42
45
63
62
61
60
58
73
72

Figure 3.18

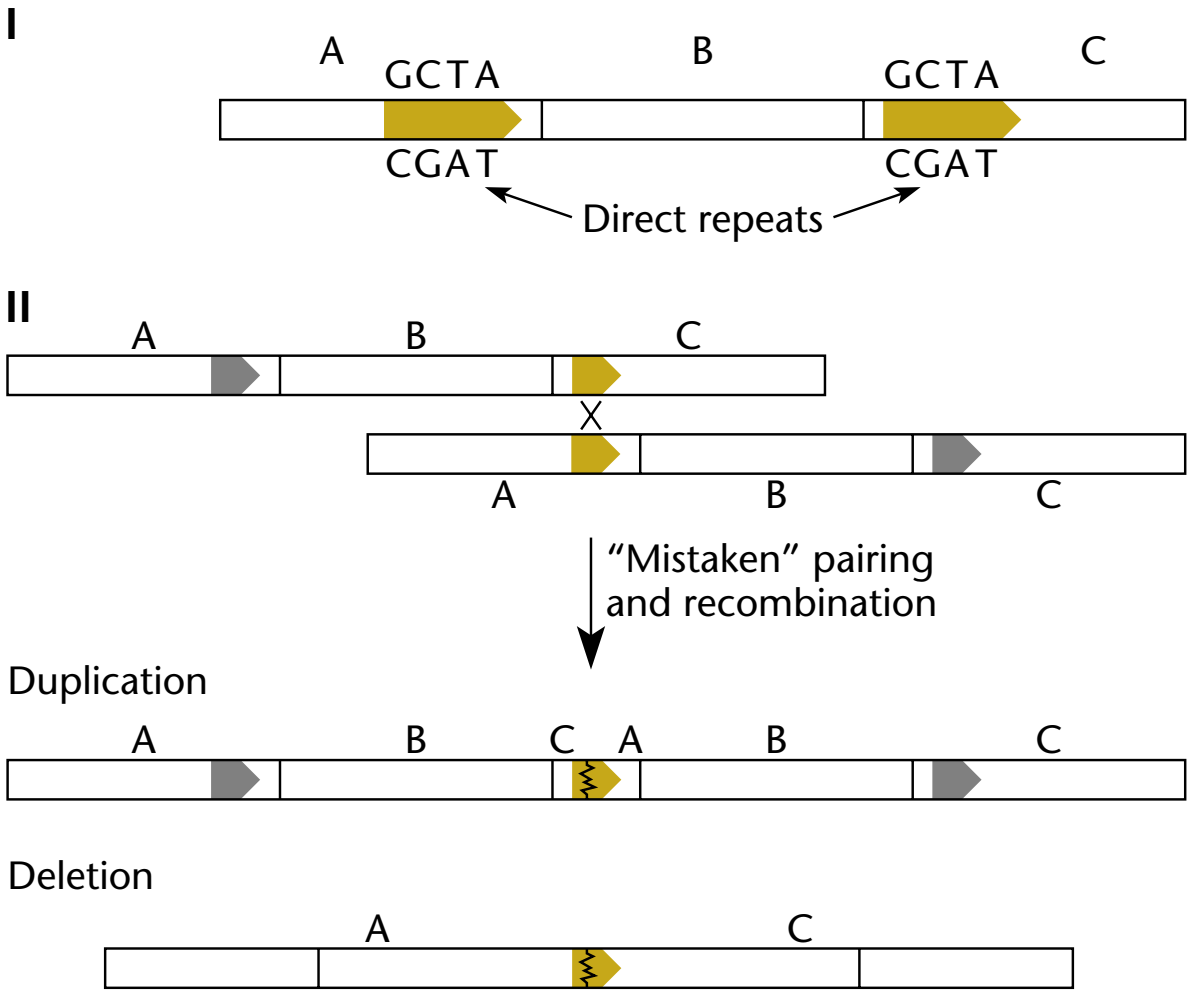
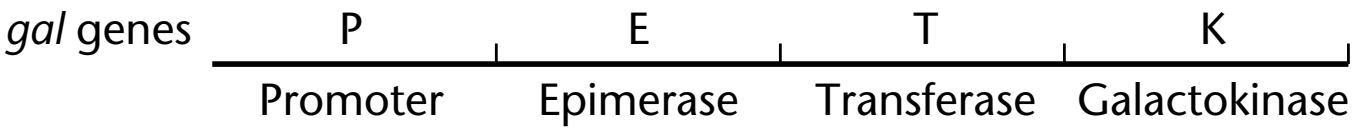

I
A
GCTA
B
GCTA
C
CGAT
CGAT
Direct repeats
II
A
B
C
X
A
B
C
"Mistaken" pairing
and recombination
Duplication
A
B
C
A
B
C
Deletion
A
C

Figure 3.19



Pathway

1 Galactose + ATP $\xrightarrow[\text{GalK}]{}$ Galactose-1-PO_4 + ADP

2 Galactose-1-PO_4 + UDPglucose $\xrightarrow[\text{GalT}]{}$ UDPgalactose + glucose

3 UDPgalactose $\xrightarrow[\text{GalE}]{}$ UDPglucose

Figure 3.20

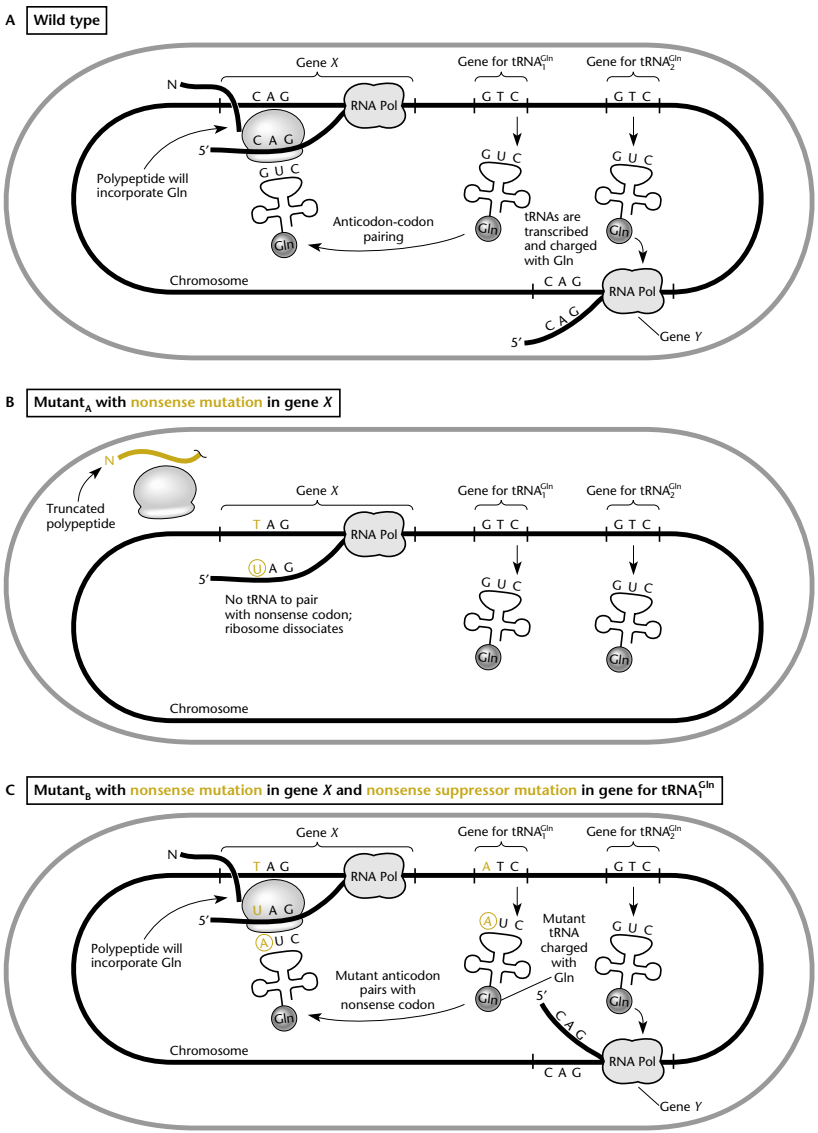

A Wild type
Gene X
Gene for tRNA$_1^{Gln}$
Gene for tRNA$_2^{Gln}$
N
C A G
RNA Pol
G T C
C A G
5′
G U C
Polypeptide will incorporate Gln
Anticodon-codon pairing
Gln
tRNAs are transcribed and charged with Gln
Chromosome
C A G
RNA Pol
C A G
5′
Gene Y
B Mutant$_A$ with nonsense mutation in gene X
N
Truncated polypeptide
Gene X
Gene for tRNA$_1^{Gln}$
Gene for tRNA$_2^{Gln}$
T A G
RNA Pol
G T C
5′
U A G
No tRNA to pair with nonsense codon; ribosome dissociates
G U C
Gln
Chromosome
C Mutant$_B$ with nonsense mutation in gene X and nonsense suppressor mutation in gene for tRNA$_1^{Gln}$
Gene X
Gene for tRNA$_1^{Gln}$
Gene for tRNA$_2^{Gln}$
N
T A G
RNA Pol
A T C
G T C
U A G
5′
A U C
Polypeptide will incorporate Gln
Mutant anticodon pairs with nonsense codon
Gln
Mutant tRNA charged with Gln
G U C
Chromosome
5′
C A G
C A G
RNA Pol
Gene Y

Table 3.4

TABLE 3.4 Some *E. coli* nonsense suppressor tRNAs

Suppressor name	tRNA	Anticodon change	Suppressor type
supE	$tRNA^{Gln}$	CUG-CUA	Amber
supF	$tRNA^{Tyr}$	GUA-CUA	Amber
supB	$tRNA^{Gln}$	UUG-UUA	Ochre/amber
supL	$tRNA^{Lys}$	UUU-UUA	Ochre/amber

Figure 3.21

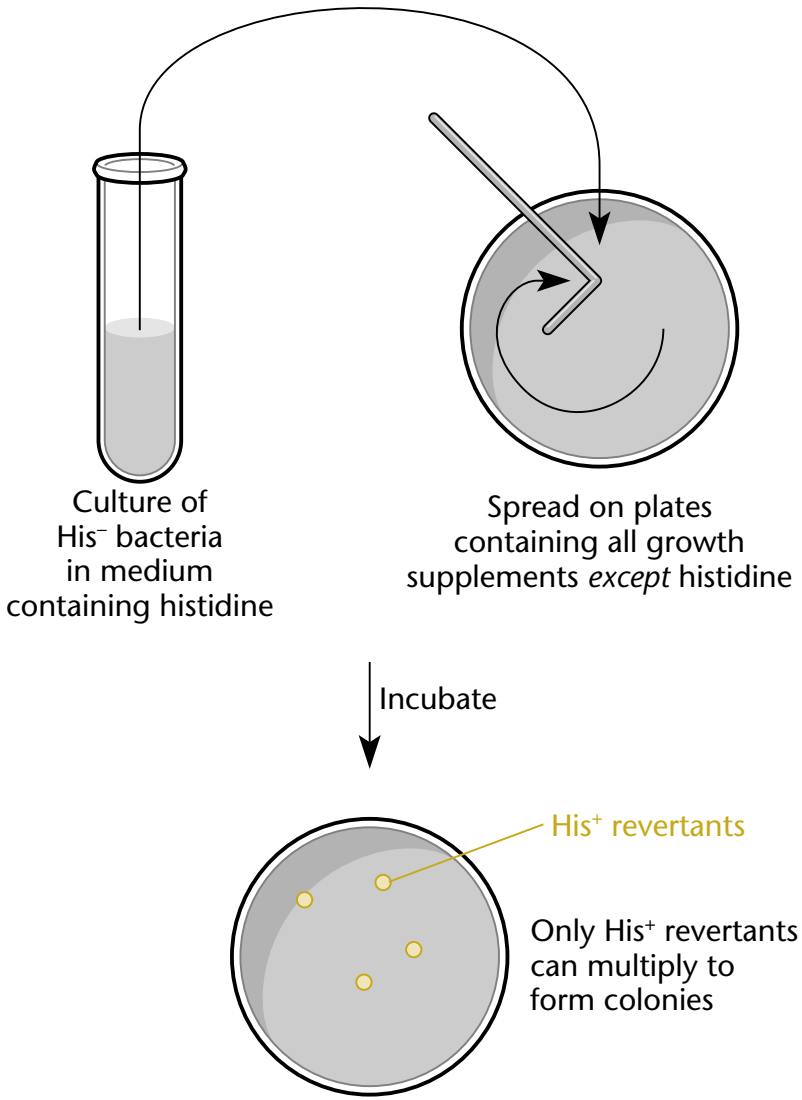

Culture of
His⁻ bacteria
in medium
containing histidine
Spread on plates
containing all growth
supplements *except* histidine
Incubate
His⁺ revertants
Only His⁺ revertants
can multiply to
form colonies

Figure 3.22

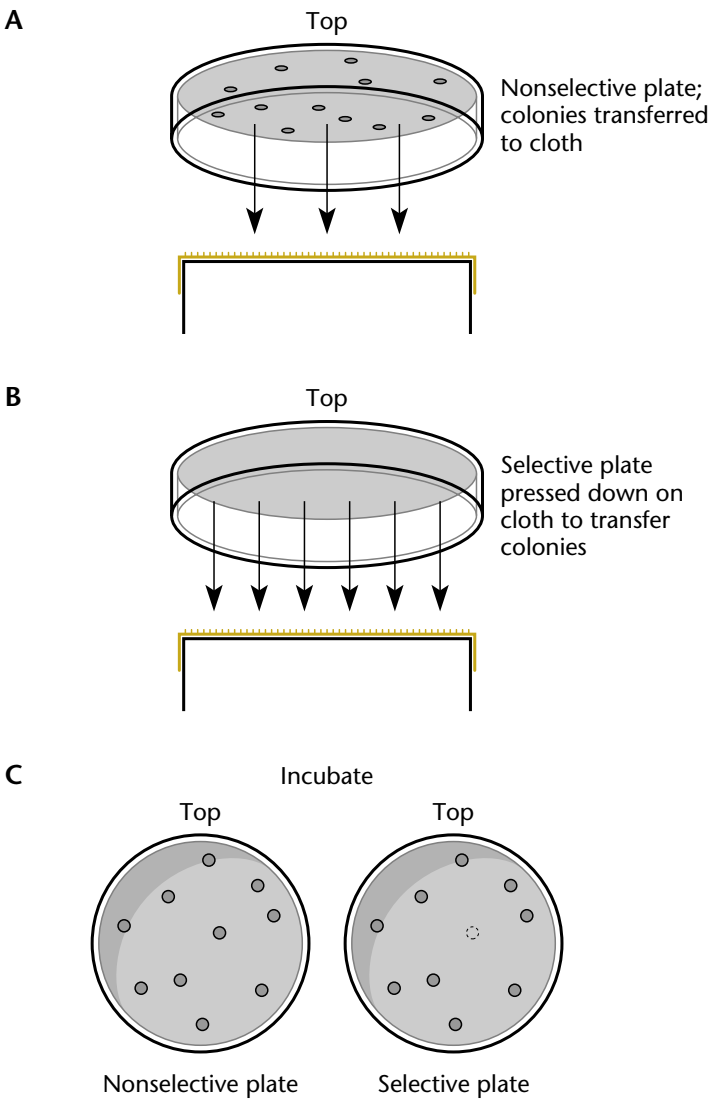

A
Top
Nonselective plate; colonies transferred to cloth
B
Top
Selective plate pressed down on cloth to transfer colonies
C
Incubate
Top
Top
Nonselective plate
Selective plate

Figure 3.23

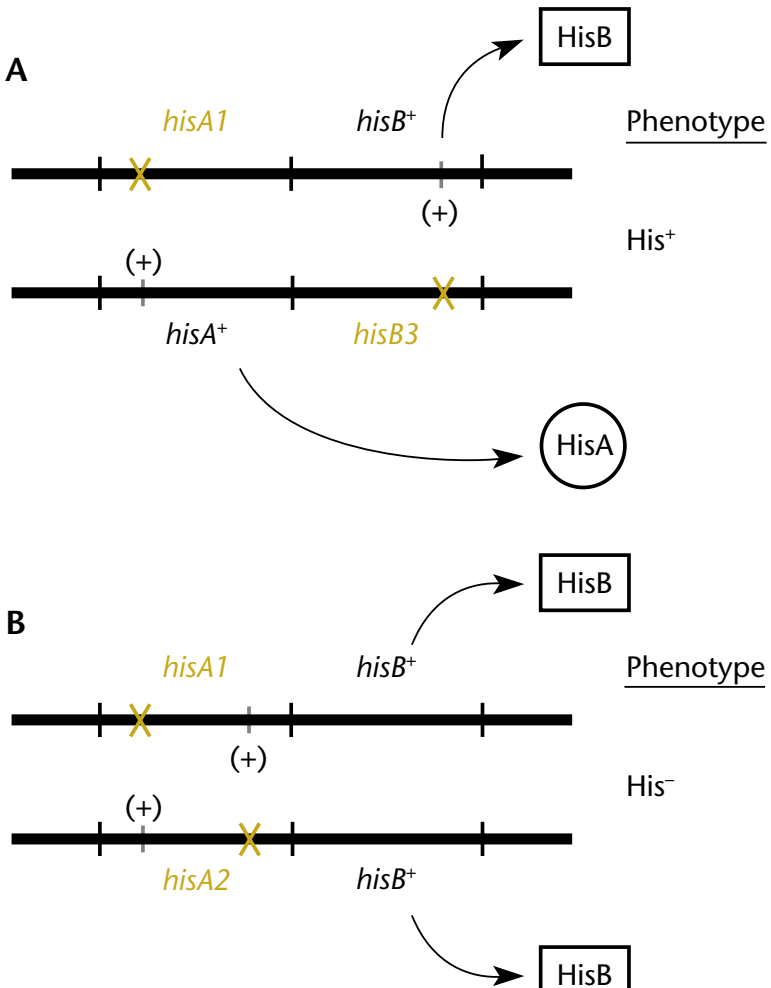

A
HisB
hisA1
hisB+
Phenotype
(+)
His+
(+)
hisA+
hisB3
HisA
B
HisB
hisA1
hisB+
Phenotype
(+)
His−
(+)
hisA2
hisB+
HisB

Table 3.5

TABLE 3.5	Interpretation of complementation tests
Test result	**Possible explanations**
x and y complement	Mutations are in different genes Intragenic complementation has occurred[a]
x and y do not complement	Mutations are in the same gene One of the mutations is dominant One of the mutations affects a regulatory site or is polar

[a]See the text for an explanation of intragenic complementation. This is a less likely explanation than the mutations being in different genes.

Figure 3.24

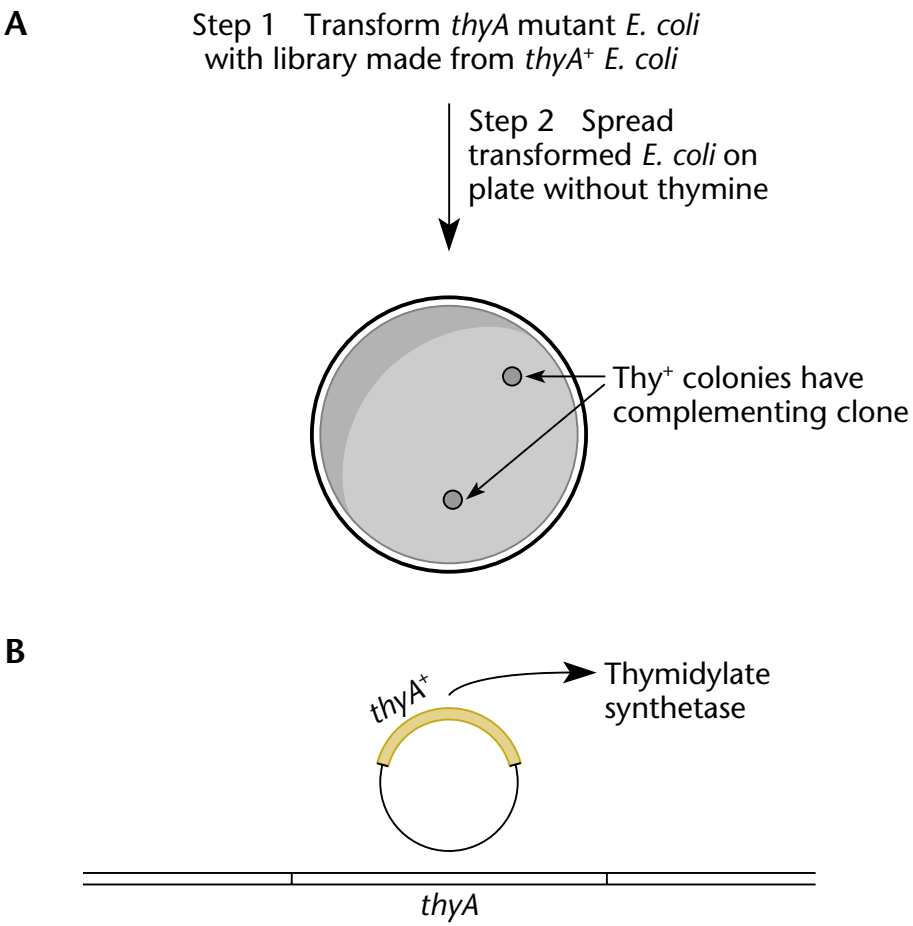

A
Step 1 Transform thyA mutant E. coli with library made from thyA+ E. coli
Step 2 Spread transformed E. coli on plate without thymine
Thy+ colonies have complementing clone
B
thyA+
Thymidylate synthetase
thyA

Figure 3.25

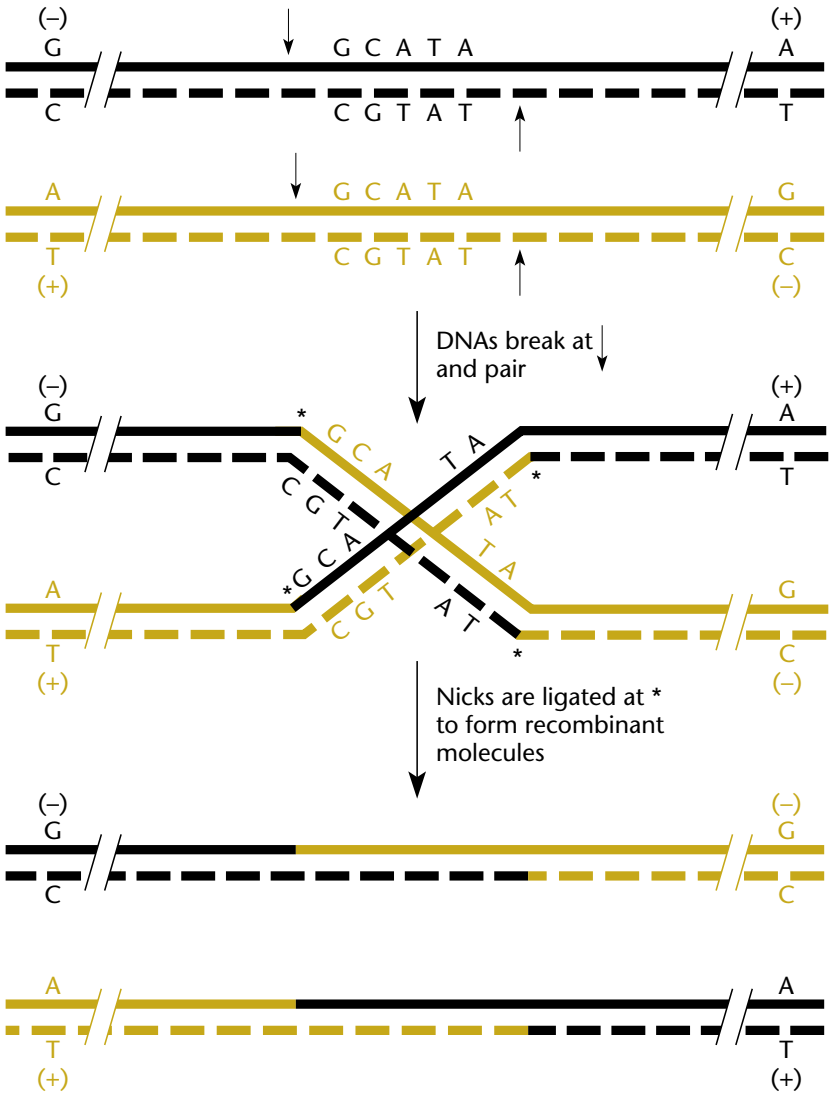

(–)
G
C
G C A T A
C G T A T
(+)
A
T
A
T
(+)
G C A T A
C G T A T
G
C
(–)
DNAs break at
and pair
(–)
G
C
(+)
A
T
*
G C A
C G T A
*G C
T A
A T
T A
T
G
C
A
T
(+)
G
C
(–)
Nicks are ligated at *
to form recombinant
molecules
(–)
G
C
(–)
G
C
A
T
(+)
A
T
(+)

Figure 3.26

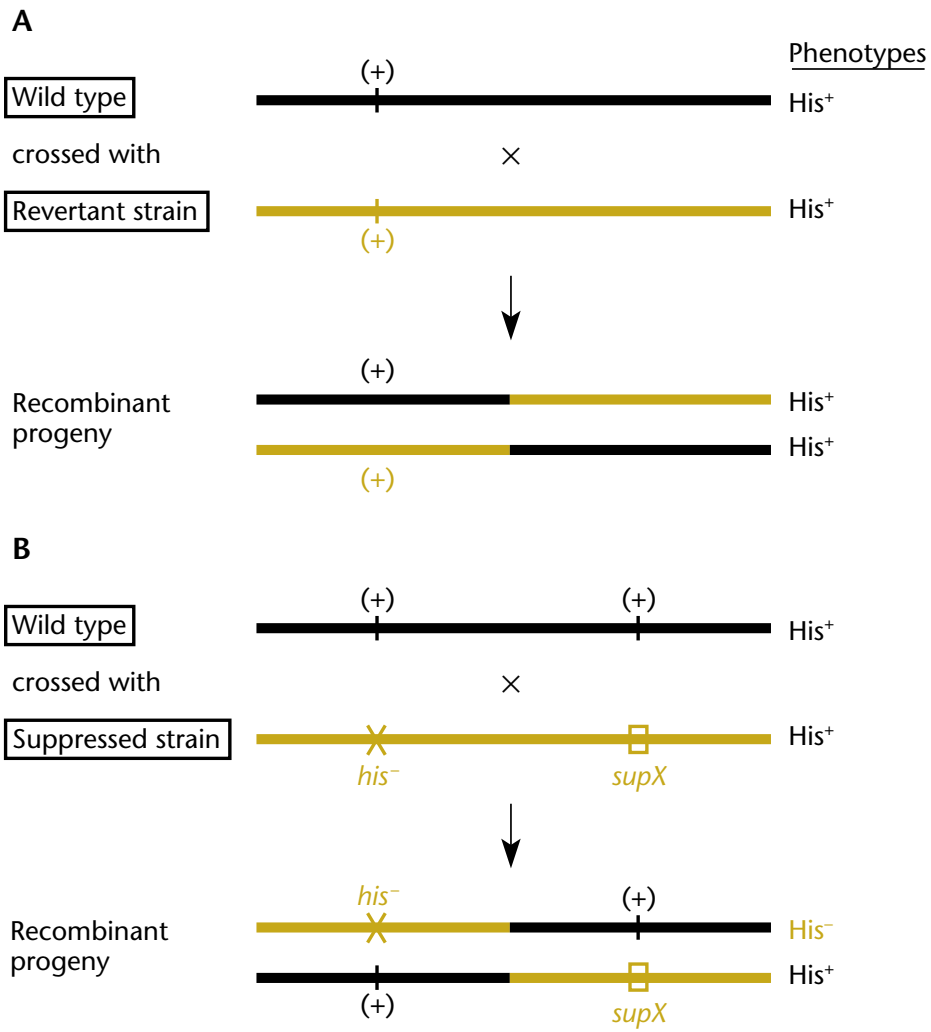

A
Phenotypes
Wild type
(+)
His+
crossed with
×
Revertant strain
His+
(+)
Recombinant progeny
(+)
His+
His+
(+)
B
Wild type
(+)
(+)
His+
crossed with
×
Suppressed strain
His+
his−
supX
his−
(+)
Recombinant progeny
His−
His+
(+)
supX

Figure 3.27

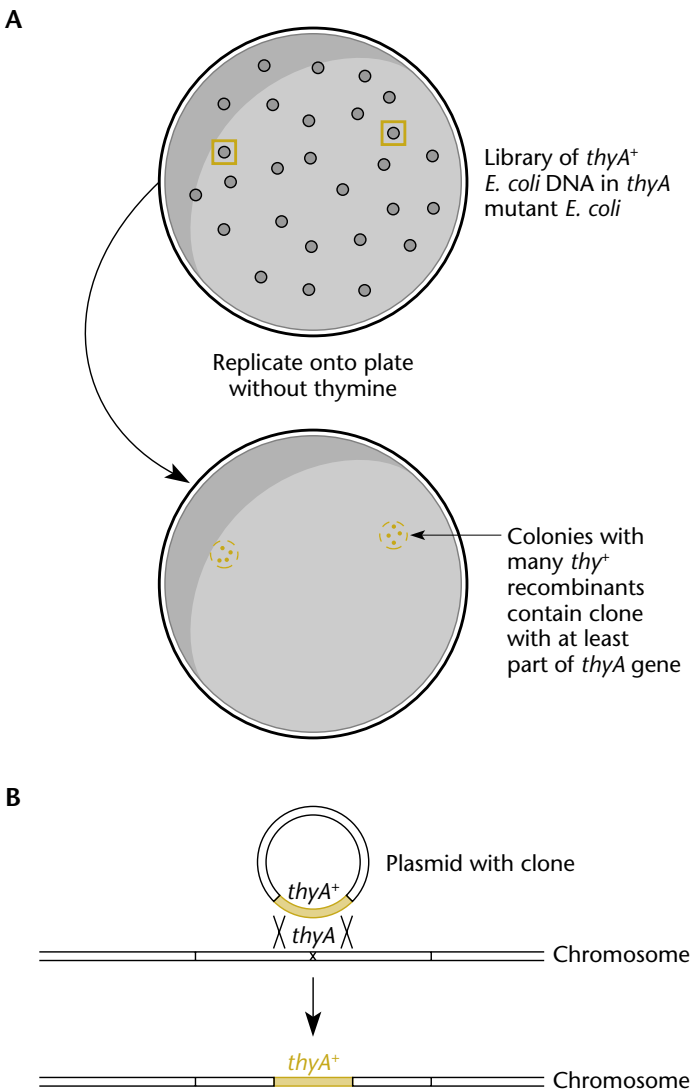

A
Library of thyA+ E. coli DNA in thyA mutant E. coli
Replicate onto plate without thymine
Colonies with many thy+ recombinants contain clone with at least part of thyA gene
B
Plasmid with clone
thyA+
thyA
Chromosome
thyA+
Chromosome

Figure 3.28

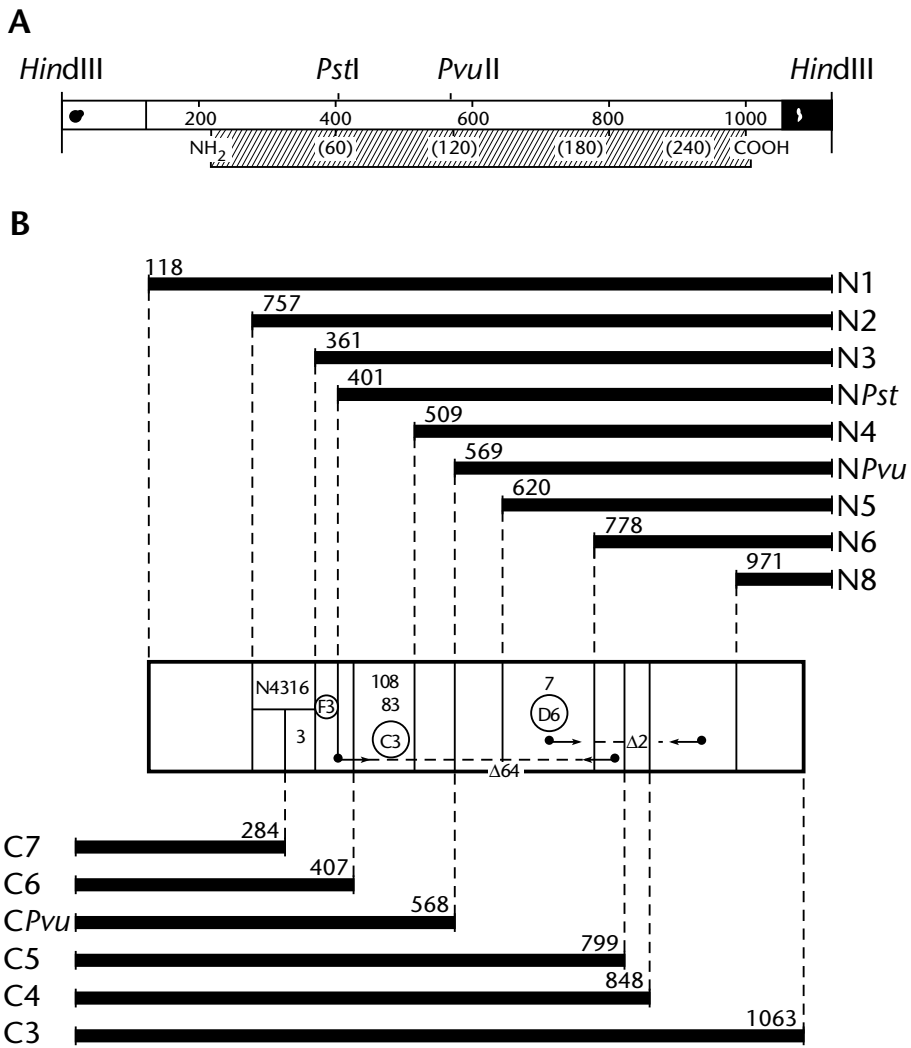

A
HindIII
PstI
PvuII
HindIII
200
400
600
800
1000
NH2
(60)
(120)
(180)
(240)
COOH
B
118
N1
757
N2
361
N3
401
NPst
509
N4
569
NPvu
620
N5
778
N6
971
N8
N4316
F3
3
108
83
C3
7
D6
Δ2
Δ64
284
C7
407
C6
568
CPvu
799
C5
848
C4
1063
C3

Figure 3.29

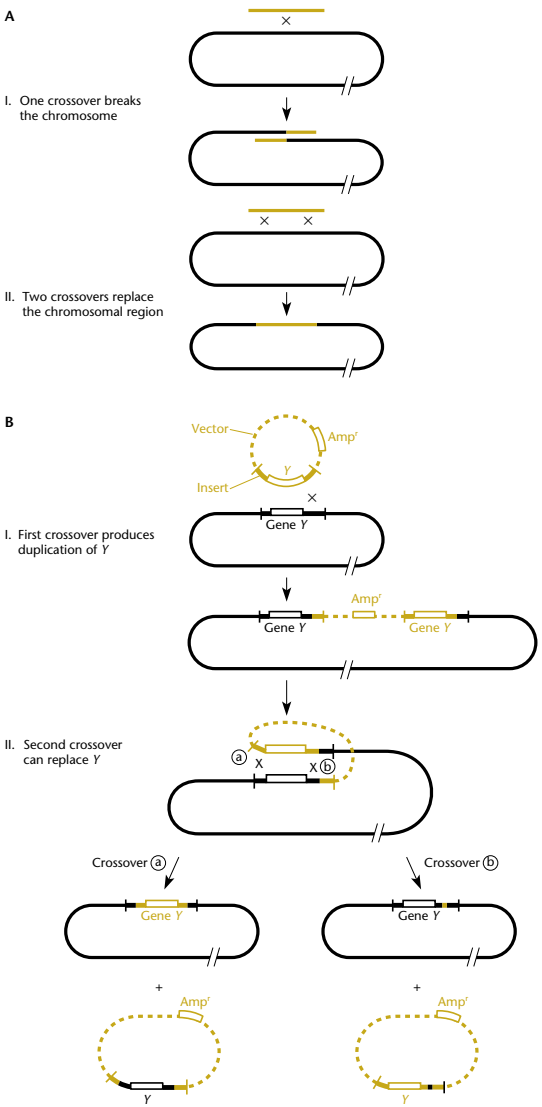

A
I. One crossover breaks the chromosome
II. Two crossovers replace the chromosomal region
B
Vector
Amp^r
Y
Insert
I. First crossover produces duplication of Y
Gene Y
Amp^r
Gene Y
II. Second crossover can replace Y
a
X
X
b
Crossover a
Crossover b
Gene Y
Gene Y
+
+
Amp^r
Amp^r
Y
Y

Figure 3.30

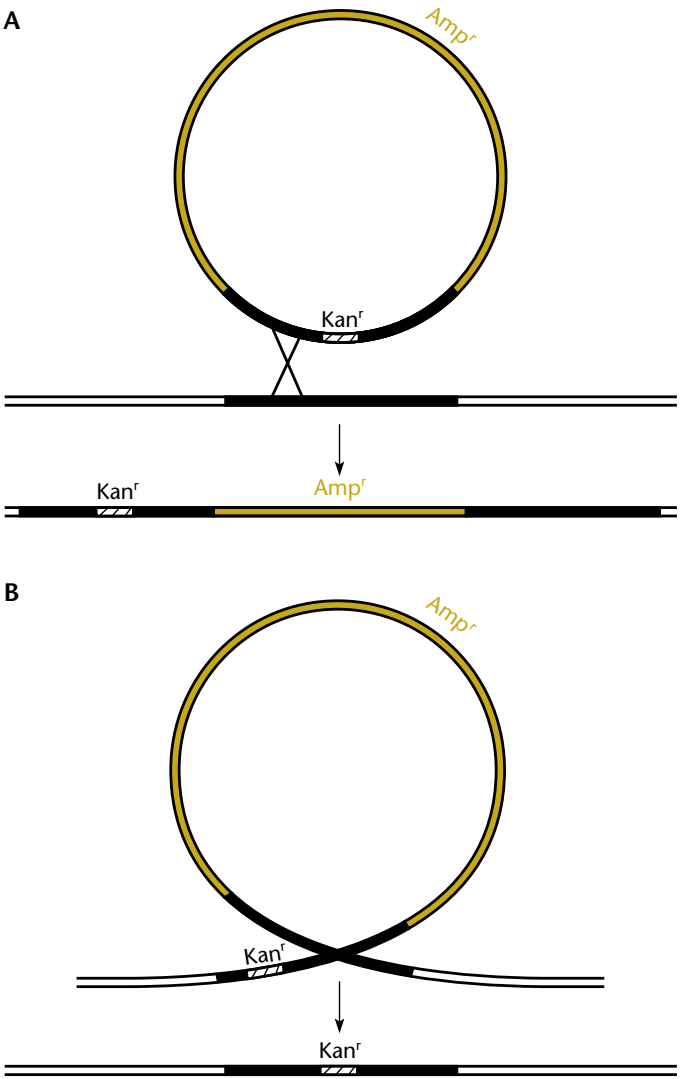
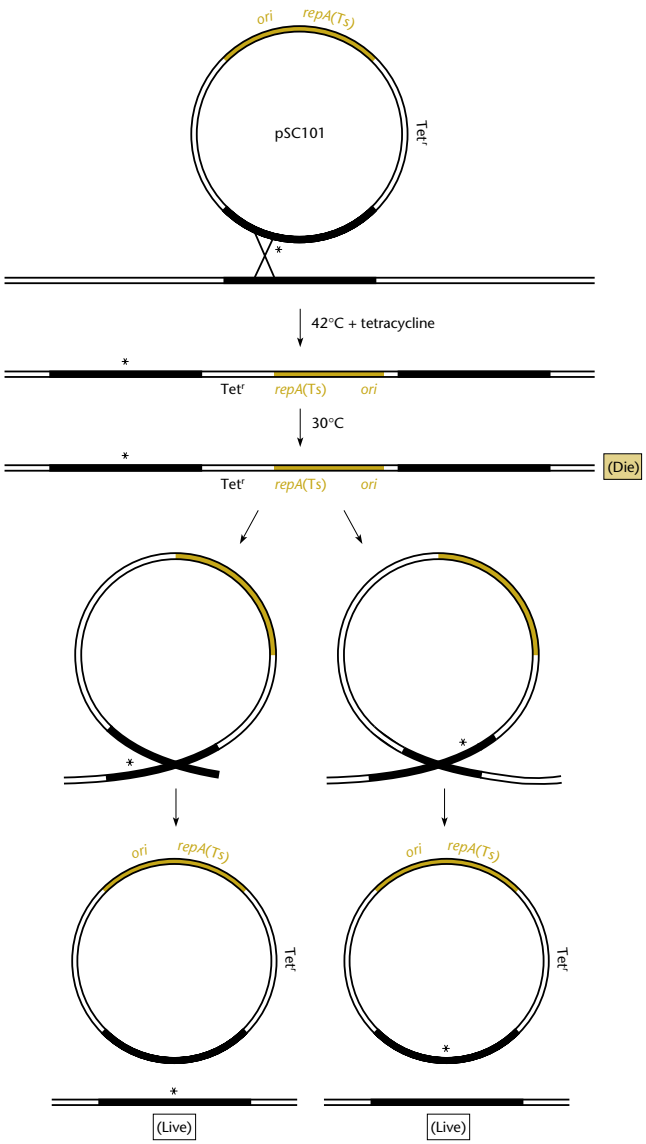


Figure 3.31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