Microbial Genetics

By
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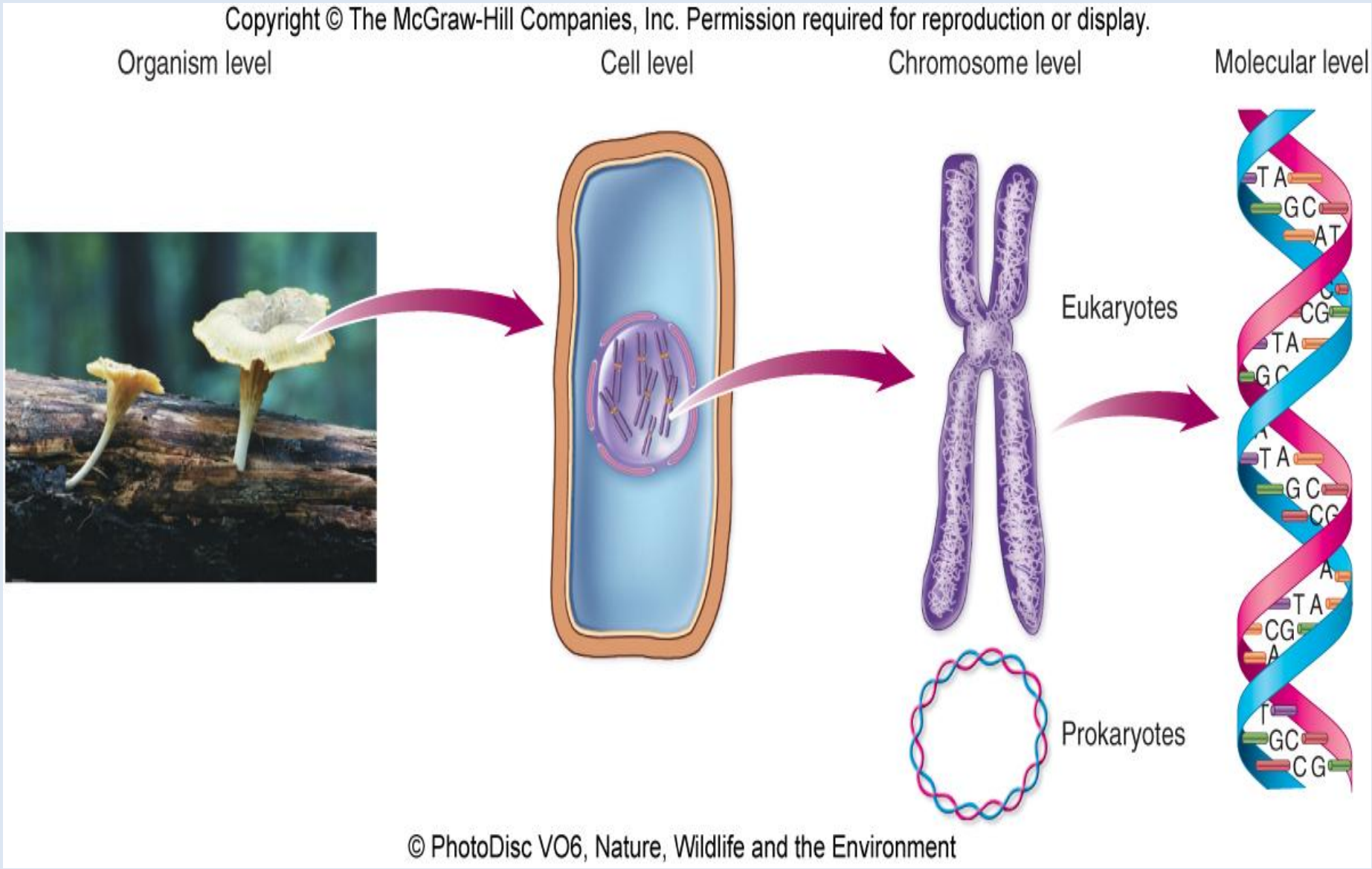
Introduction to Genetics and Genes

- **Genetics**: the study of the inheritance (**heredity**) of living things
  - Transmission of traits from parent to offspring
  - Expression and variation of those traits
  - The structure and function of the genetic material
  - How this material changes
- Takes place on several levels: organismal, chromosomal, molecular

Microbial Genetics



The Nature of the Genetic Material

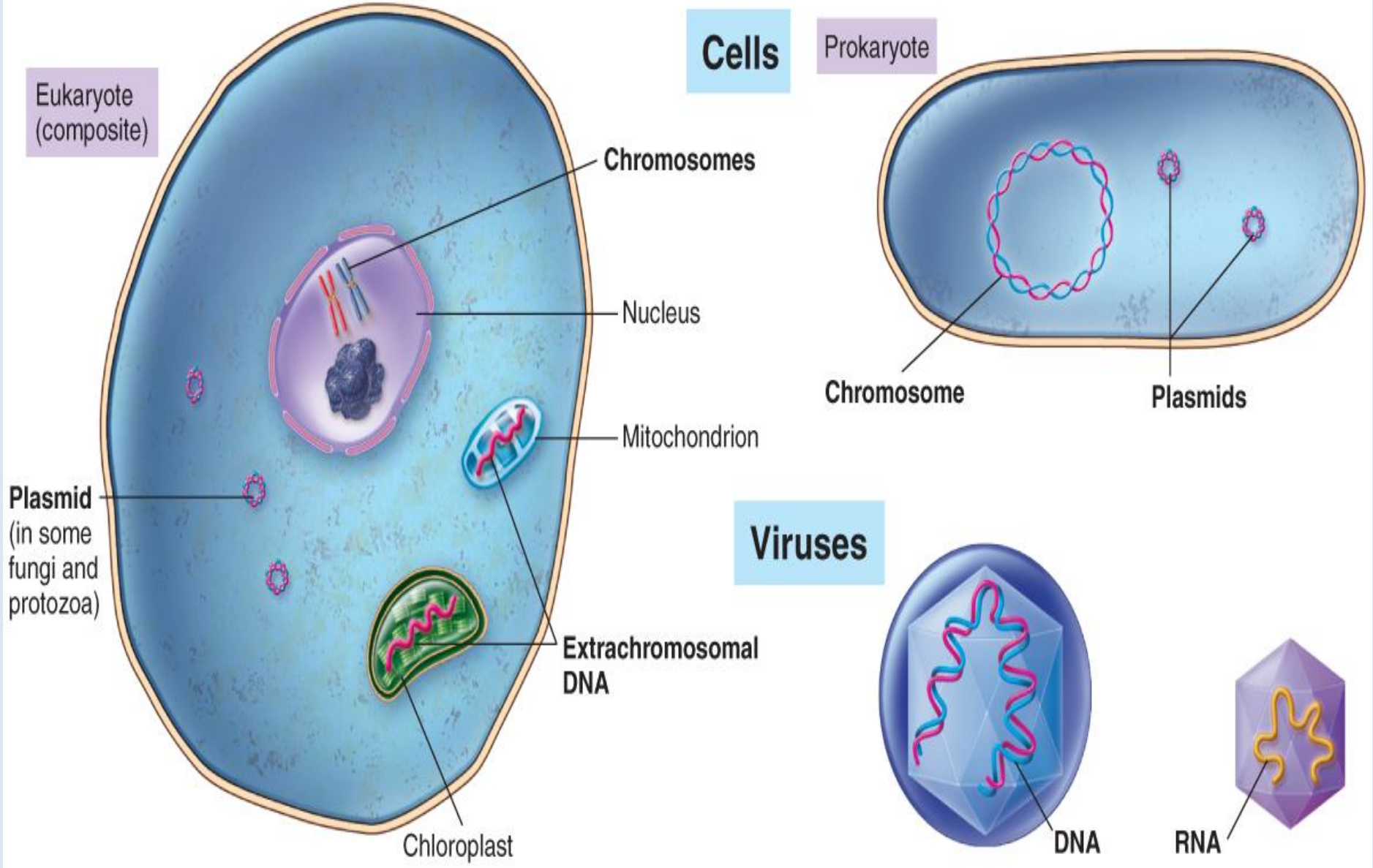
- Must be able to self-replicate
- Must be accurately duplicated and separated from each daughter cell

The Levels of Structure and Function of the Genome

- Genome
- Chromosome
- Gene

Genome

- The sum total of genetic material of a cell
- Mostly in chromosomes
- Can appear in nonchromosomal sites as well
- In cells- exclusively DNA
- In viruses- can be either DNA or RNA



Chromosome

- A discrete cellular structure composed of a neatly packed DNA molecule

- Eukaryotic chromosomes
  - DNA molecule tightly wound around histone proteins
  - Located in the nucleus
  - Vary in number from a few to hundreds
  - Can occur in pairs (diploid) or singles (haploid)
  - Appear linear

- Bacterial chromosomes
  - Condensed and secured by means of histone-like proteins
  - Single, circular chromosome

Gene

- A certain segment of DNA that contains the necessary code to make a protein or RNA molecule
- Structural genes: code for proteins or code for RNA
- Regulatory genes: control gene expression
- Sum of all genes is an organism's **genotype**
- The expression of the genotype creates traits which make up the **phenotype.** Some genes may not be expressed in the phenotype.
- All organisms contain more genes in their genotype than are manifested as a phenotype at a given time

The Size and Packaging of Genomes

- Vary greatly in size
  - Smallest viruses- 4 or 5 genes
  - *Escherichia coli*- 4,288 genes
  - Human cell- 20,000 to 25,000 genes

- The stretched-out DNA can be 1,000 times or more longer than the cell

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The DNA Code: A Simple Yet Profound Message

- 1953: James Watson and Francis Crick
  - Discovered DNA is a gigantic molecule
  - A type of nucleic acid
  - With two strands combined into a double helix

General Structure of DNA

- Basic unit: **nucleotide**
  - **Phosphate**
  - **Deoxyribose sugar**
  - **Nitrogenous base**

Nucleotides

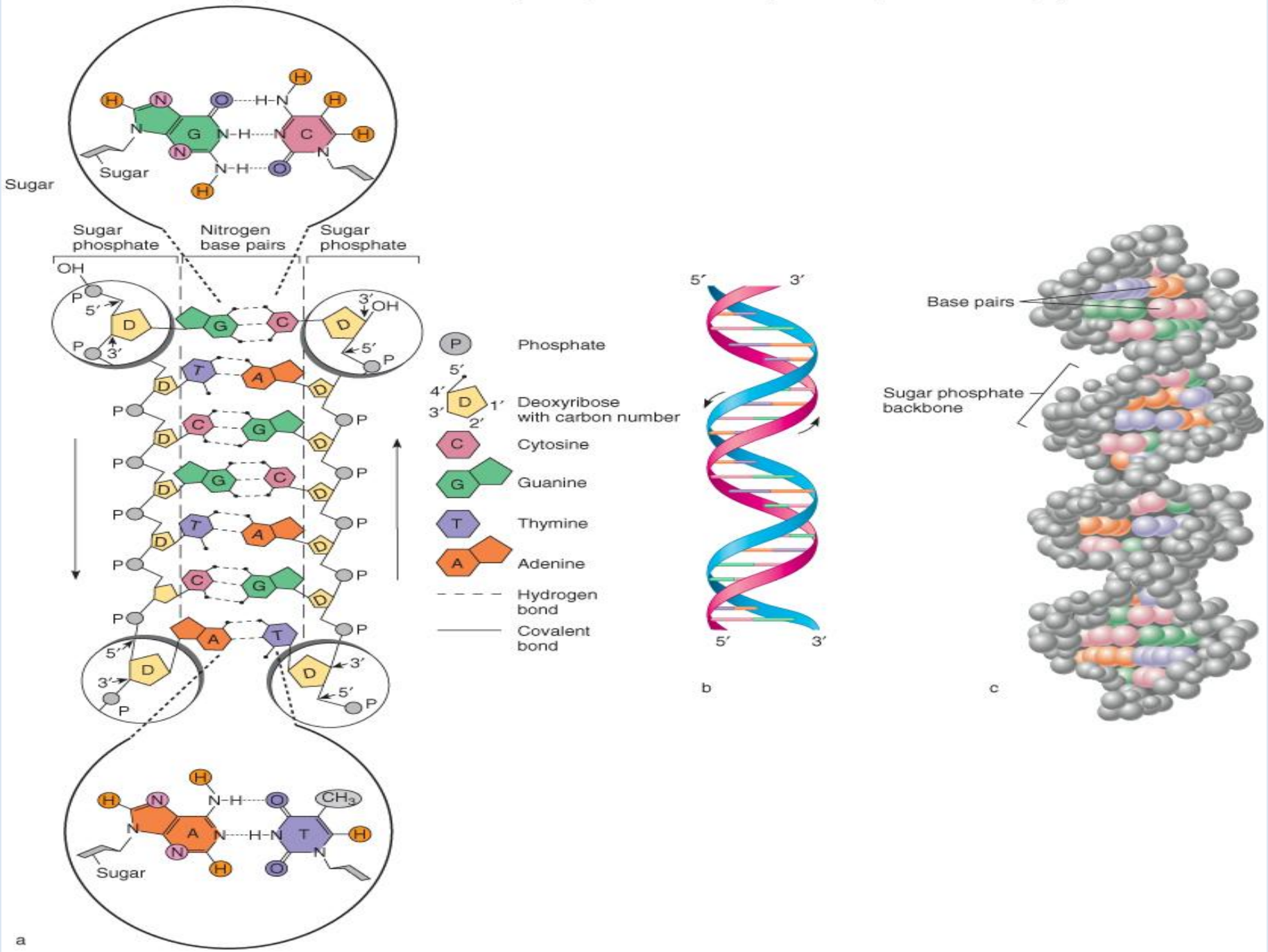
- Covalently bond to form a sugar-phosphate linkage- the backbone of each strand
- Each sugar attaches to two phosphates
- One bond is to the 5’ carbon on deoxyribose
- The other is to the 3’ carbon

Nitrogenous Bases

- **Purines** and **pyrimidines**
- Attach by covalent bonds at the 1’ position of the sugar
- Span the center of the molecule and pair with complementary bases from the other strands
- The paired bases are joined by hydrogen bonds
  - Easily broken
  - Allow the molecule to be “unzipped”
- **Adenine** always pairs with **thymine**
- **Guanine** always pairs with **cytosine**

Antiparallel Arrangment

- One side of the helix runs in the opposite direction of the other- antiparallel
- One helix runs from 5’ to 3’ direction
- The other runs from 3’ to 5’



The Significance of DNA Structure

- Arrangement of nitrogenous bases
  - Maintains the code during reproduction (conservative replication of DNA)
  - Provides variety

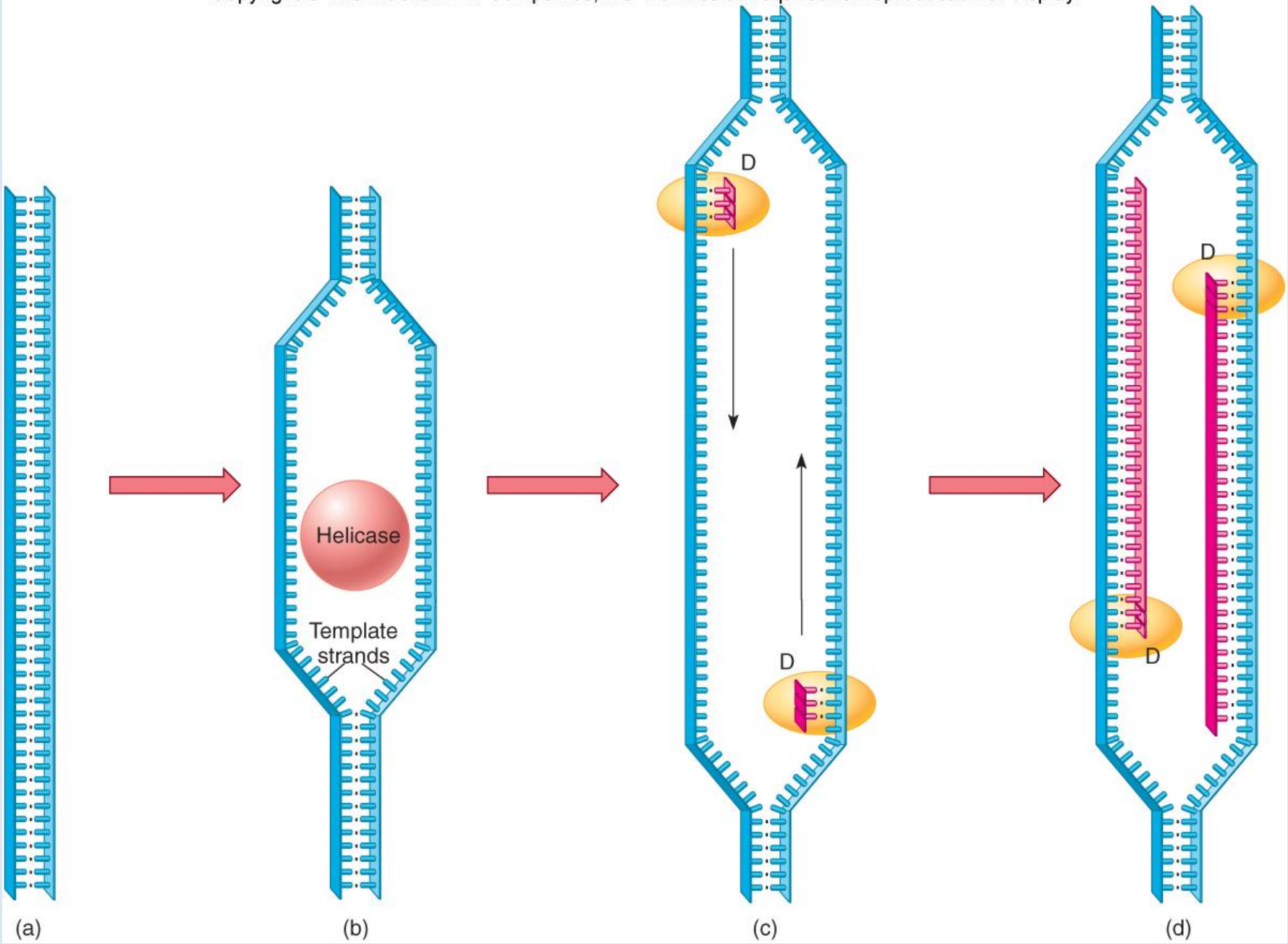


Figure 9.5

DNA Replication: Preserving the Code and Passing it On

- The process of the genetic code duplicated and passed on to each offspring
- Must be completed during a single generation time

The Overall Replication Process

- Requires the actions of 30 different enzymes
  - Separate the strands
  - Copy its template
  - Produce two new daughter molecules

TABLE 9.1 Some Enzymes Involved in DNA Replication and Their Functions

Enzyme	Function
Helicase	Unzipping the DNA helix
Primase	Synthesizing an RNA primer
DNA polymerase III	Adding bases to the new DNA chain; proofreading the chain for mistakes
DNA polymerase I	Removing primer, closing gaps, repairing mismatches
Ligase	Final binding of nicks in DNA during synthesis and repair
Gyrase	Supercoiling

Semiconservative Replication

- Each daughter molecule is identical to the parent in composition, but only one strand is completely new
- The parent DNA molecule uncoils
- The hydrogen bonds between the base pairs are unzipped
  - Separates the two strands
  - Exposes the nucleotide sequence of each strand to serve as templates
- Two new strands are synthesized by attachment of the correct complementary nucleotides to each single-stranded template

Refinements and Details of Replication

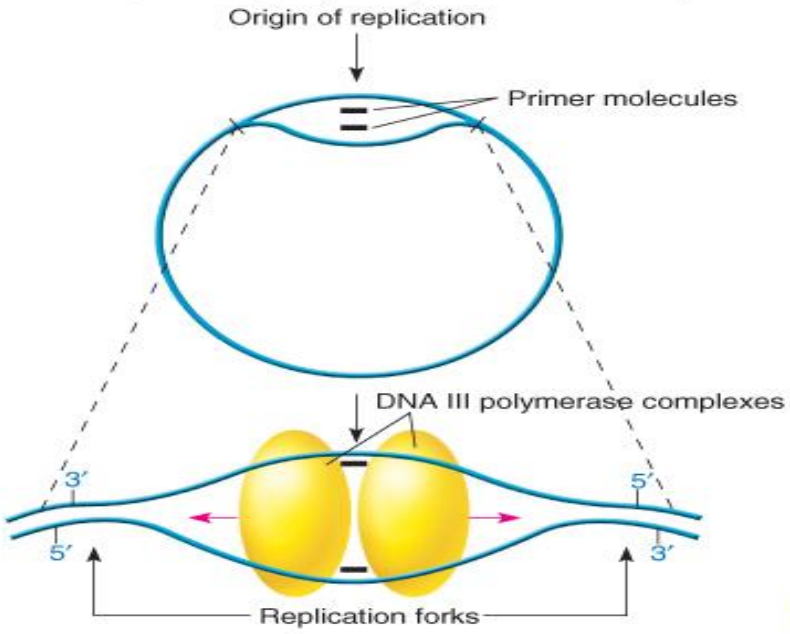
- Origin of replication
  - Short sequence
  - Rich in A and T
  - Held together by only two H bonds rather than three
  - Less energy is required to separate the two strands

- Helicases bind to the DNA at the origin
  - Untwist the helix
  - Break the hydrogen bonds
  - Results in two separate strands

DNA Polymerase III

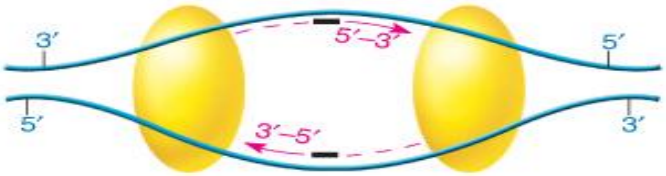
- Synthesizes a new daughter strand using the parental strand as a template

- The process depends on several other enzymes as well, but key points about DNA polymerase III:
  - Nucleotides that need to be read by DNA polymerase III are buried in the double helix- so the DNA must first be unwound and the two strands separated
  - DNA polymerase III is unable to begin synthesizing a chain of nucleotides but can only continue to add nucleotides to an already existing chain
  - DNA polymerase III always reads the original strand from 3” to 5”
  - DNA polymerase III can only add nucleotides in one direction, so a new strand is always synthesized from 5’ to 3’

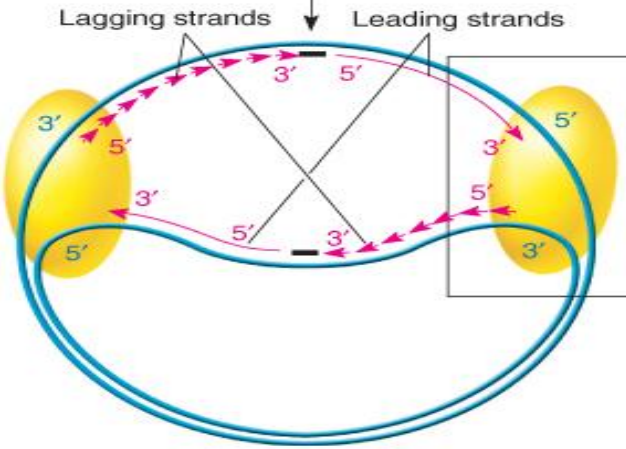


1. Replication origin. Short RNA primers are positioned to start replication.

2. Strands separate; two polymerase complexes attach at origin. Arrows indicate direction of replication.

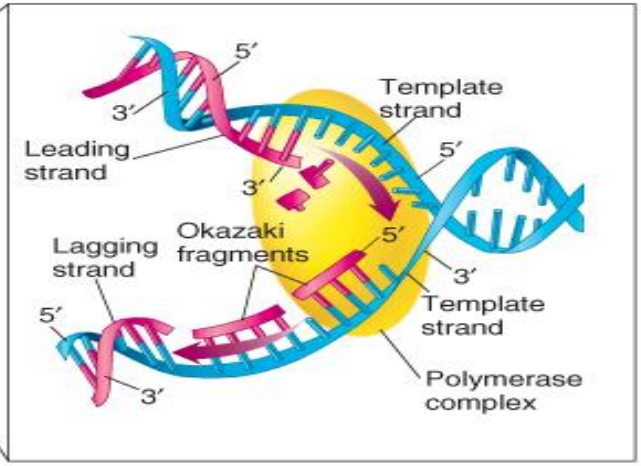


3. At primer sequence, each polymerase complex synthesizes two strands at the replication forks.



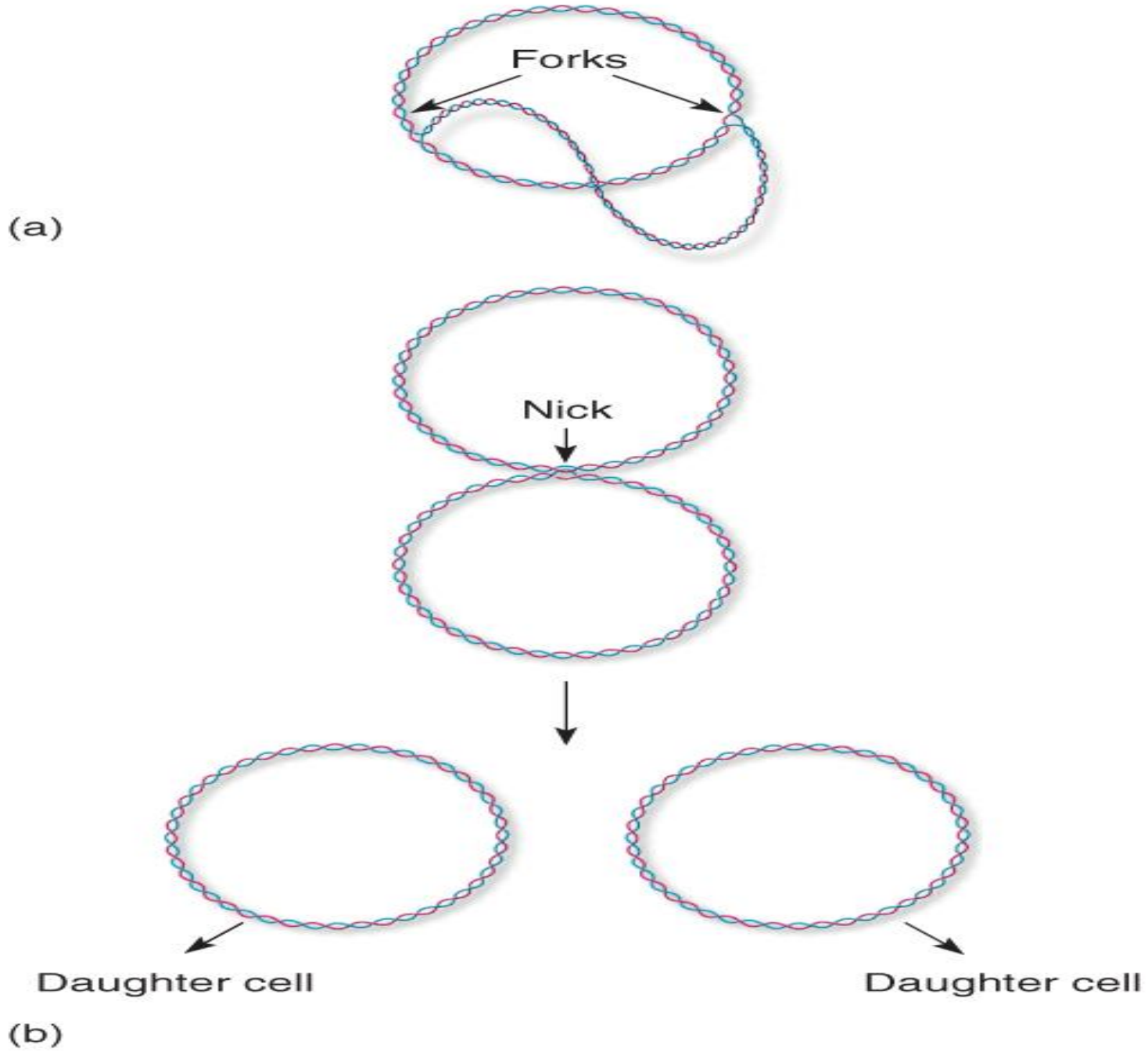
4. Since DNA polymerase acts only in the 5′ to 3′ direction, it forms a continuous leading strand from that orientation. The lagging strand, which orients 3′ to 5′, must be made backward in short sections, 5′ to 3′, which are later linked together. Note that the numbers refer to the direction of synthesis of the *new* strand (red).

5. In actuality, a loop forms in the lagging strand so that the portion of the lagging strand undergoing replication is oriented in the same direction as the leading strand (i.e., both strands are parallel). This allows the polymerase complex to move along both strands in the 5′ to 3′ prime direction at the same time, synthesizing both strands simultaneously. Details of the process are seen in the inset, although, for purposes of clarity, the loop is not shown.



Elongation and Termination of the Daughter Molecules

- As replication proceeds, the newly produced double strand loops down
- DNA polymerase I removes RNA primers and replaces them with DNA
- When the forks come full circle and meet, ligases move along the lagging strand
  - Begin initial linking of the fragments
  - Complete synthesis and separation of the two circular daughter molecules



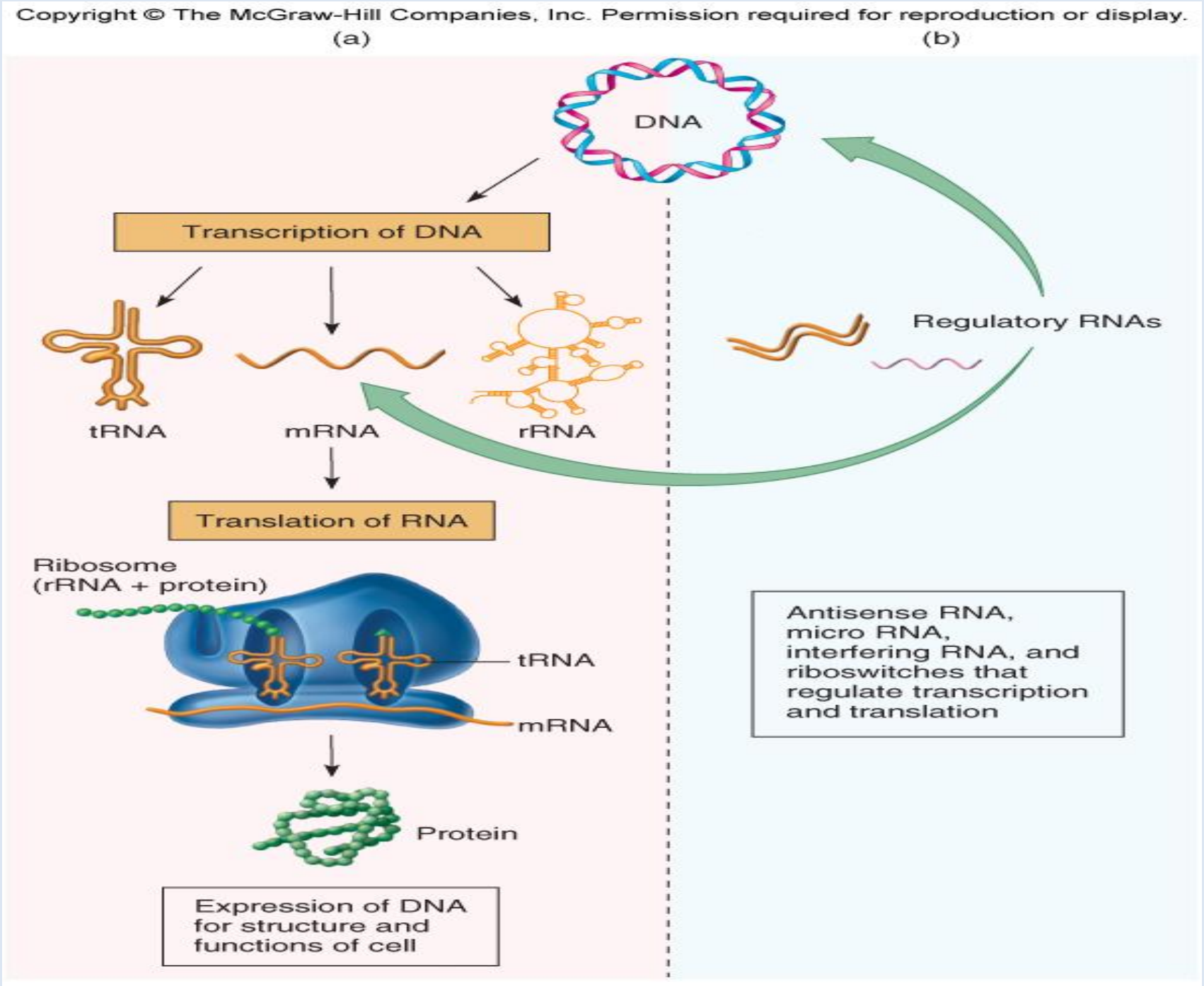
- Occasionally an incorrect base is added to the growing chain
- Most are corrected
- If not corrected, result in mutations

- DNA polymerase III can detect incorrect, unmatching bases, excise them, and replace them with the correct base

- DNA polymerase I can also proofread and repair

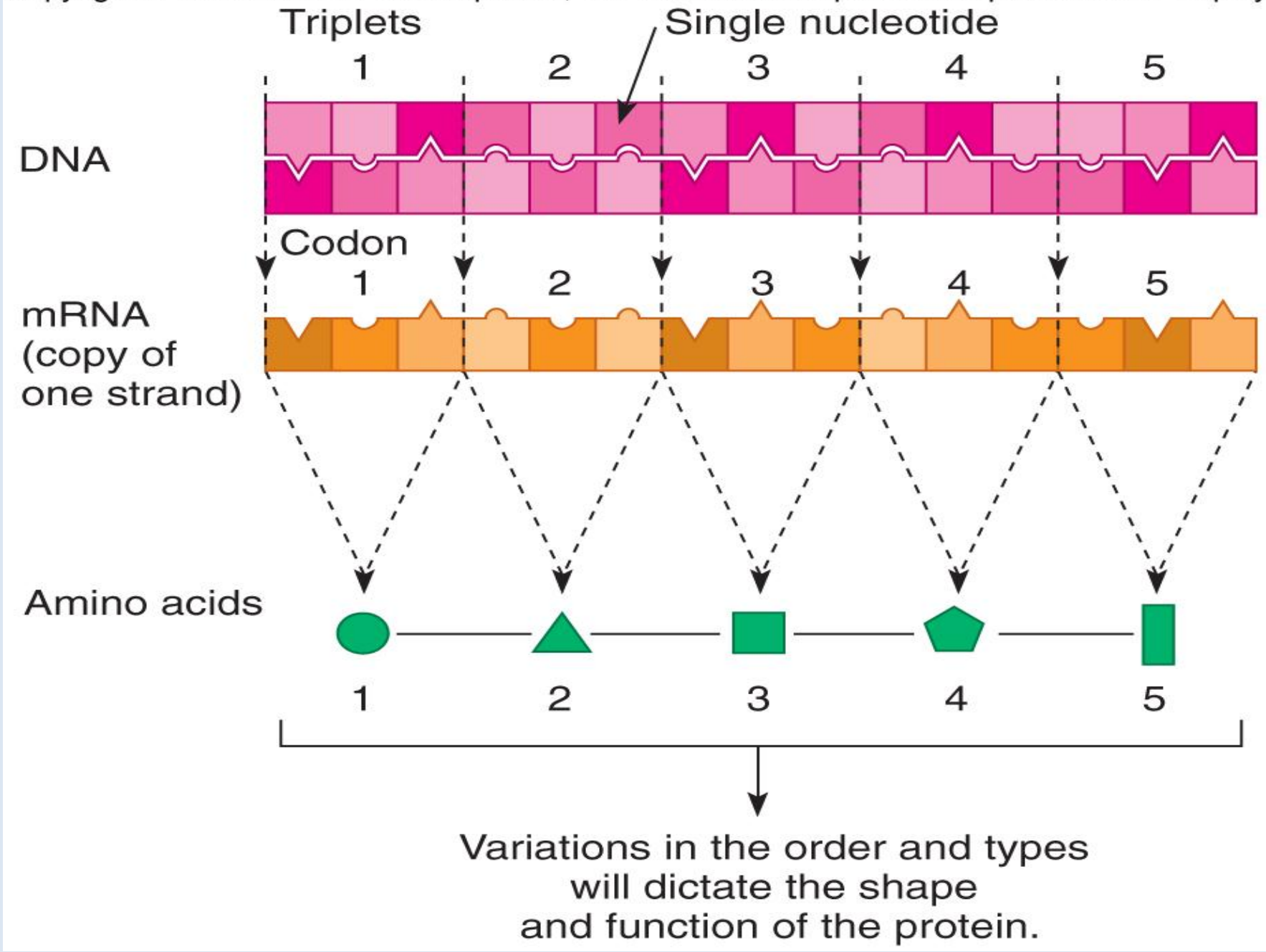
9.2 Applications of the DNA Code: Transcription and Translation

- Central dogma
  - Genetic information flows from DNA to RNA to protein
    - The master code of DNA is used to synthesize an RNA molecule (**transcription**)
    - The information in the RNA is used to produce proteins (**translation**)
    - Exceptions: RNA viruses and retroviruses
  - Recently shown to be incomplete
    - In addition to the RNA that produces protein, other RNAs are used to regulate gene function
    - Many of the genetic malfunctions that cause human disease are found in these regulatory RNA segments



The Gene-Protein Connection

- The Triplet Code and the Relationship to Proteins
  - Three consecutive bases on the DNA strand- called triplets
  - A gene differs from another in its composition of triplets
  - Each triplet represents a code for a particular amino acid
  - When the triplet code is transcribed and translated, it dictates the type and order of amino acids in a polypeptide chain
- A protein's primary structure determines its characteristic shape and function
- Proteins ultimately determine phenotype
- DNA is mainly a blueprint that tells the cell which kinds of proteins and RNAs to make and how to make them



The Major Participants in Transcription and Translation

- Number of components participate, but most prominent:
  - mRNA
  - tRNA
  - regulatory RNAs
  - ribosomes
  - several types of enzymes
  - storehouse of raw materials

- RNAs: Tools in the Cell's Assembly Line
  - RNA differs from DNA
    - Single stranded molecule
    - Helical form
    - Contains **uracil** instead of thymine
    - The sugar is ribose
  - Many functional types, from small regulatory pieces to large structural ones
  - Only mRNA is translated into a protein molecule

TABLE 9.2 Types of Ribonucleic Acid

RNA Type	Contains Codes For	Function in Cell	Translated
Messenger (mRNA)	Sequence of amino acids in protein	Carries the DNA master code to the ribosome	Yes
Transfer (tRNA)	A cloverleaf tRNA to carry amino acids	Brings amino acids to ribosome during translation	No
Ribosomal (rRNA)	Several large structural rRNA molecules	Forms the major part of a ribosome and participates in protein synthesis	No
Micro (miRNA) antisense, riboswitch, and small interfering (siRNA)	Regulatory RNAs	Regulation of gene expression and coiling of chromatin	No
Primer	An RNA that can begin DNA replication	Primes DNA	No
Ribozymes	RNA enzymes, parts of splicer enzymes	Remove introns from other RNAs in eukaryotes	No

Messenger RNA: Carrying DNA's Message

- A transcript of a structural gene or genes in the DNA
- Synthesized by the enzyme RNA polymerase
- Synthesized by a process similar to synthesis of the leading strand during DNA replication
- The message of this transcribed strand is later read as a series of triplets (**codons**)

Transfer RNA: The Key to Translation

- Also a copy of a specific region of DNA
- It is uniform in length (75-95 nucleotides long)
- Contains sequences of bases that form hydrogen bonds with complementary sections of the same tRNA strand
- At these points the molecule bends back upon itself into several hairpin loops, giving the molecule a cloverleaf structure that then folds into a complex, 3-D helix

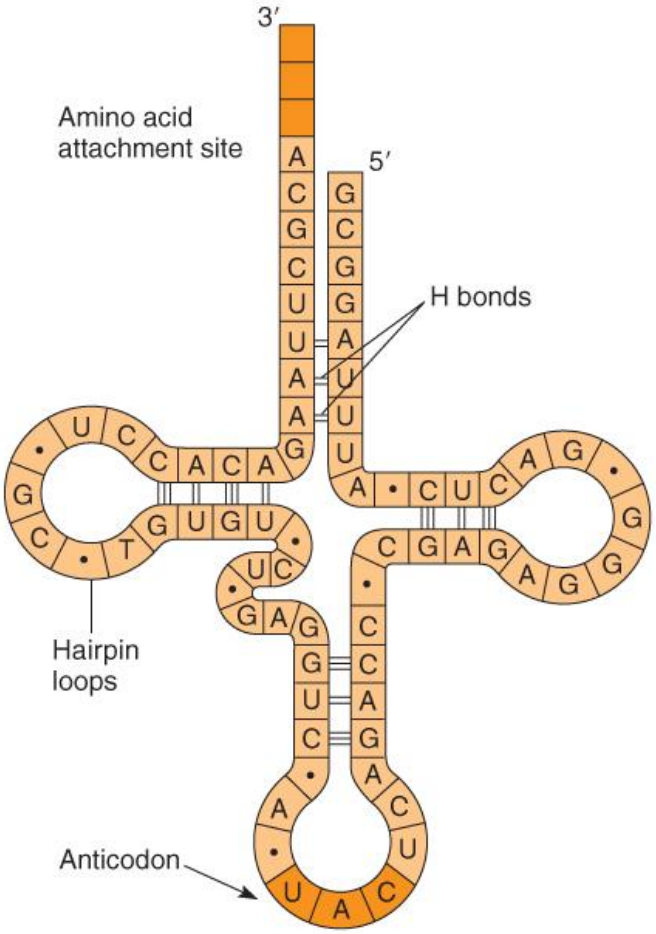
Transfer RNA: The Key to Translation cont.

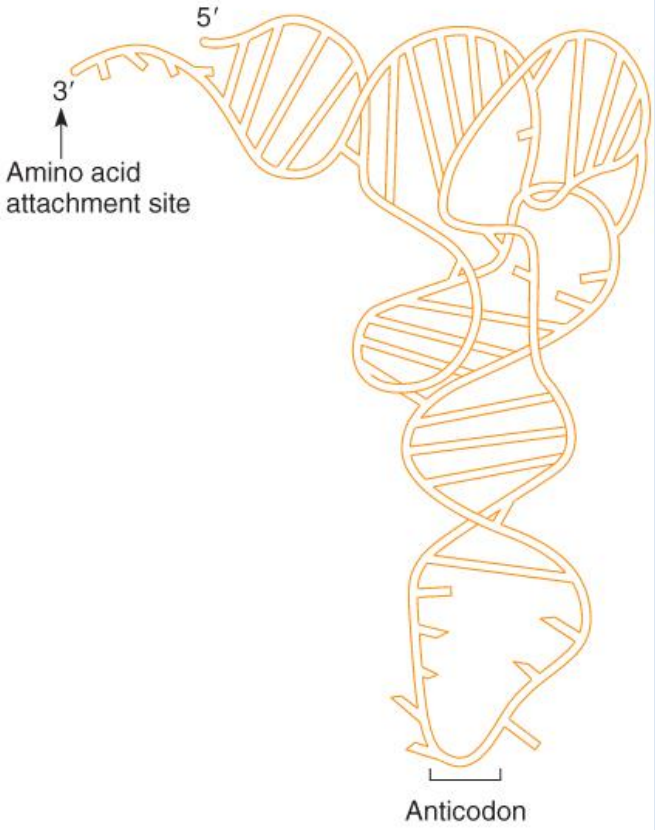
- Bottom loop of the cloverleaf exposes a triplet (the **anticodon**) that designates the specificity of the tRNA and complements mRNA's codons

- At the opposite end of the molecule is a binding site for the amino acid that is specific for that anticodon

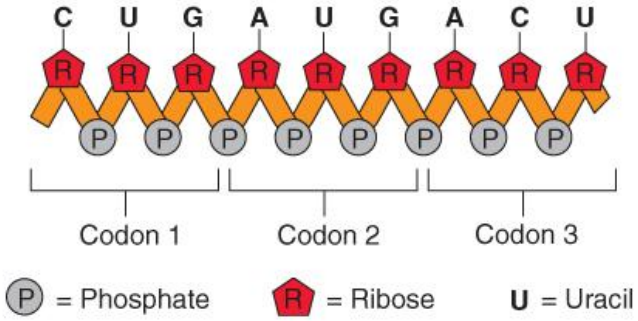
- For each of the 20 amino acids there is at least one specialized type of tRNA to carry it

(a) **Transfer RNA (tRNA).** Transfer RNA (tRNA) can loop back on itself to form intrachain hydrogen bonds. The result of the secondary structure is a cloverleaf structure, shown here in simplified form. At its bottom is an anticodon that specifies the attachment of a particular amino acid at the 3′ end. At right is a three-dimensional view of the tertiary structure of tRNA.





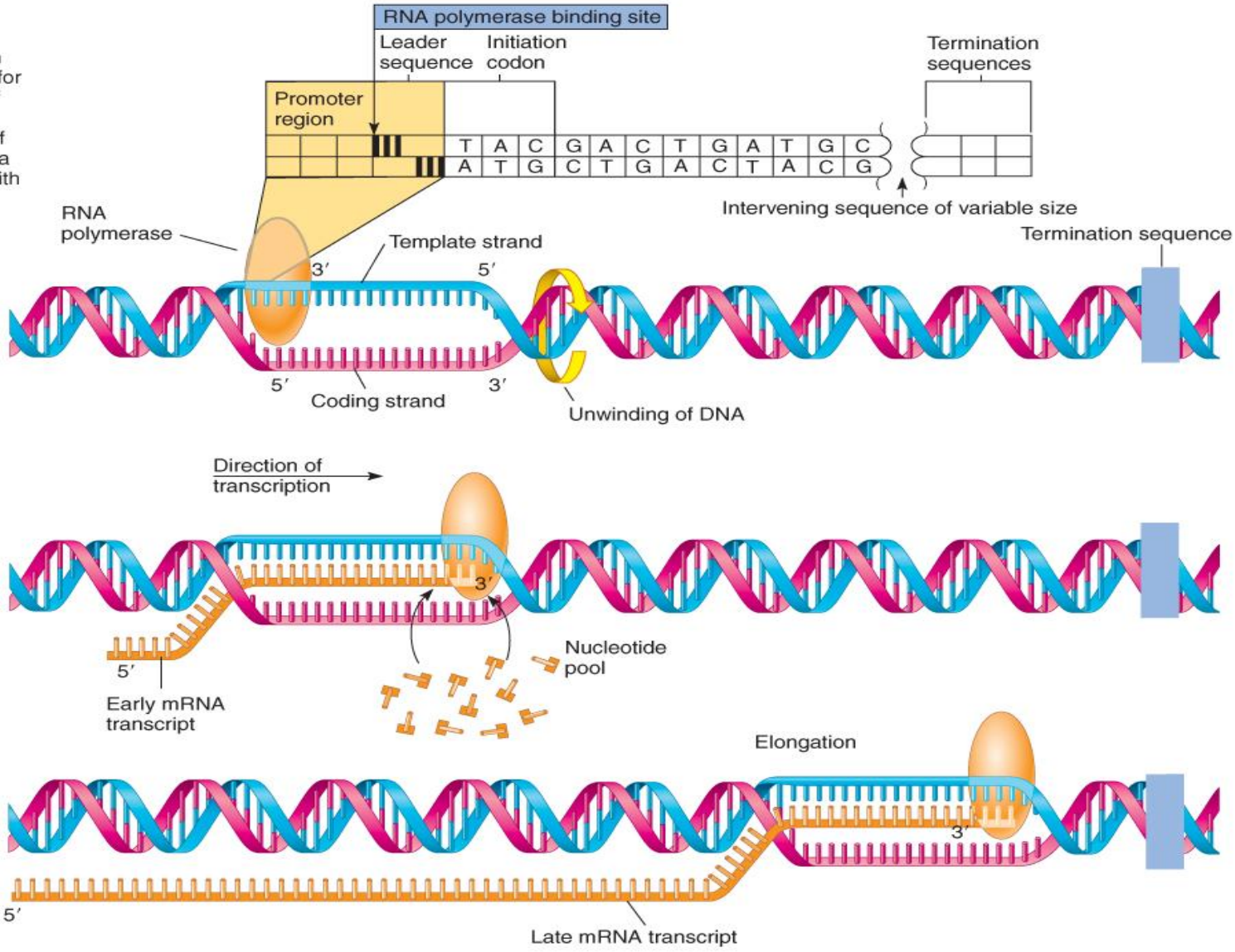
(b) **Messenger RNA (mRNA).** A short piece of messenger RNA (mRNA) illustrates the general structure of RNA: single strandedness, repeating phosphate-ribose sugar backbone attached to single nitrogen bases; use of uracil instead of thymine.



The Ribosome: A Mobile Molecular Factory for Translation

- The prokaryotic (70S) ribosome composed of tightly packed rRNA and protein
- The interactions of proteins and rRNA create the two subunits of the ribosome that engage in final translation of the genetic code
- The rRNA component of each subunit is a long polynucleotide molecule

Transcription: The First Stage of Gene Expression



1. Overall view of a gene. Each gene contains a specific promoter region and a leader sequence for guiding the beginning of transcription. This is followed by the region of the gene that codes for a polypeptide and ends with a series of terminal sequences that stop translation.
2. DNA is unwound at the promoter by RNA polymerase. Only one strand of DNA, called the template strand, is copied by the RNA polymerase. This strand runs in the 3′ to 5′ direction.
3. As the RNA polymerase moves along the strand, it adds complementary nucleotides as dictated by the DNA template, forming the single-stranded mRNA that reads in the 5′ to 3′ direction.
4. The polymerase continues transcribing until it reaches a termination site and the mRNA transcript is released for translation. Note that the section of the DNA that has been transcribed is rewound into its original configuration.

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Translation: The Second Stage of Gene Expression

- All of the elements needed to synthesize a protein are brought together on the ribosomes

- Five stages: initiation, elongation, termination, protein folding, and protein processing

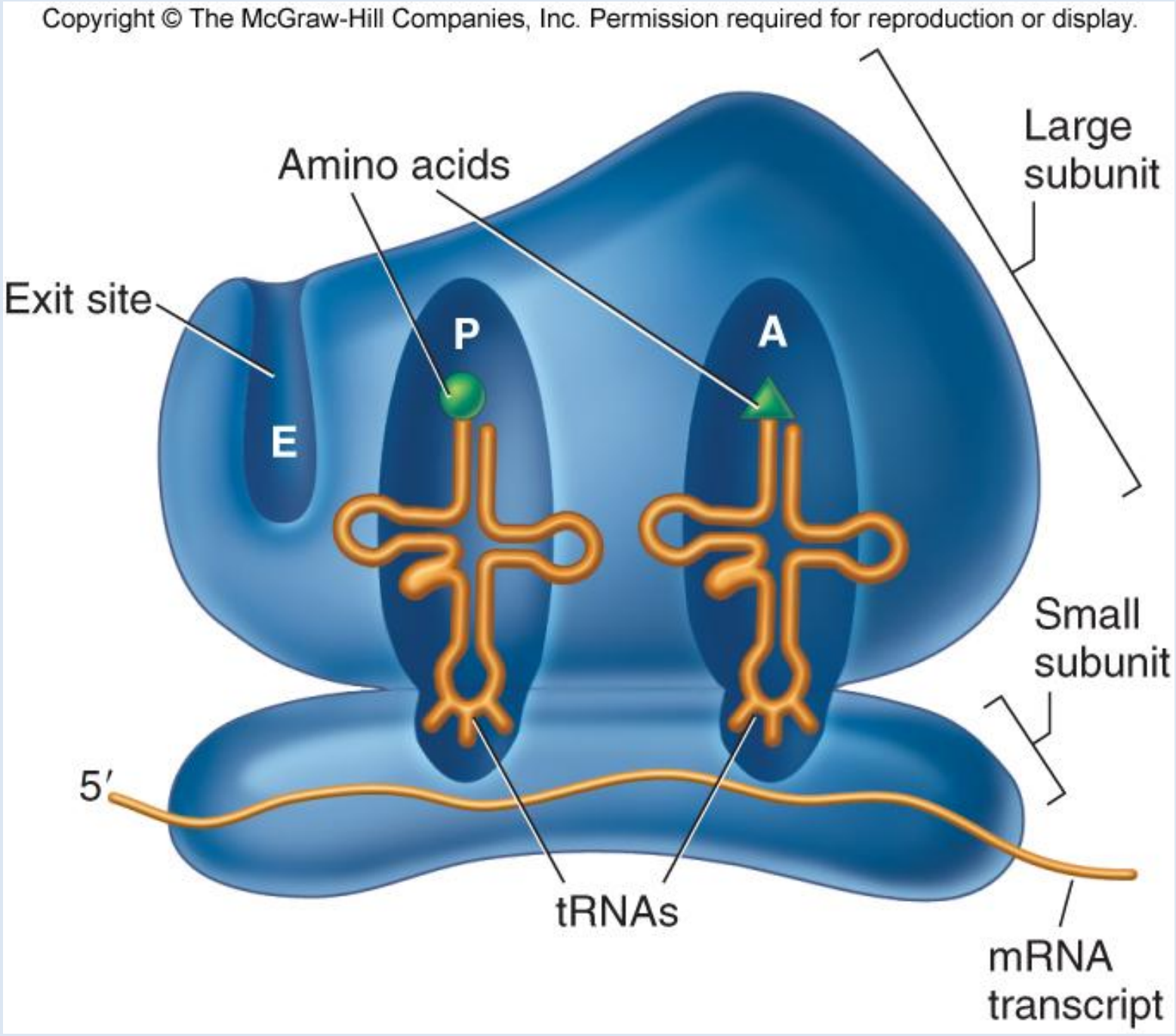


Figure 9.12

Initiation of Translation

- mRNA molecule leaves DNA transcription site
- Is transported to ribosomes in the cytoplasm
- Ribosomal subunits are specifically adapted to assembling and forming sites to hold the mRNA and tRNA's
- Prokaryotic ribosomes
  - 70s size
    - 50s subunit
    - 30s subunit
- Eukaryotic ribosomes
  - 80s
    - 60s subunit
    - 40s subunit

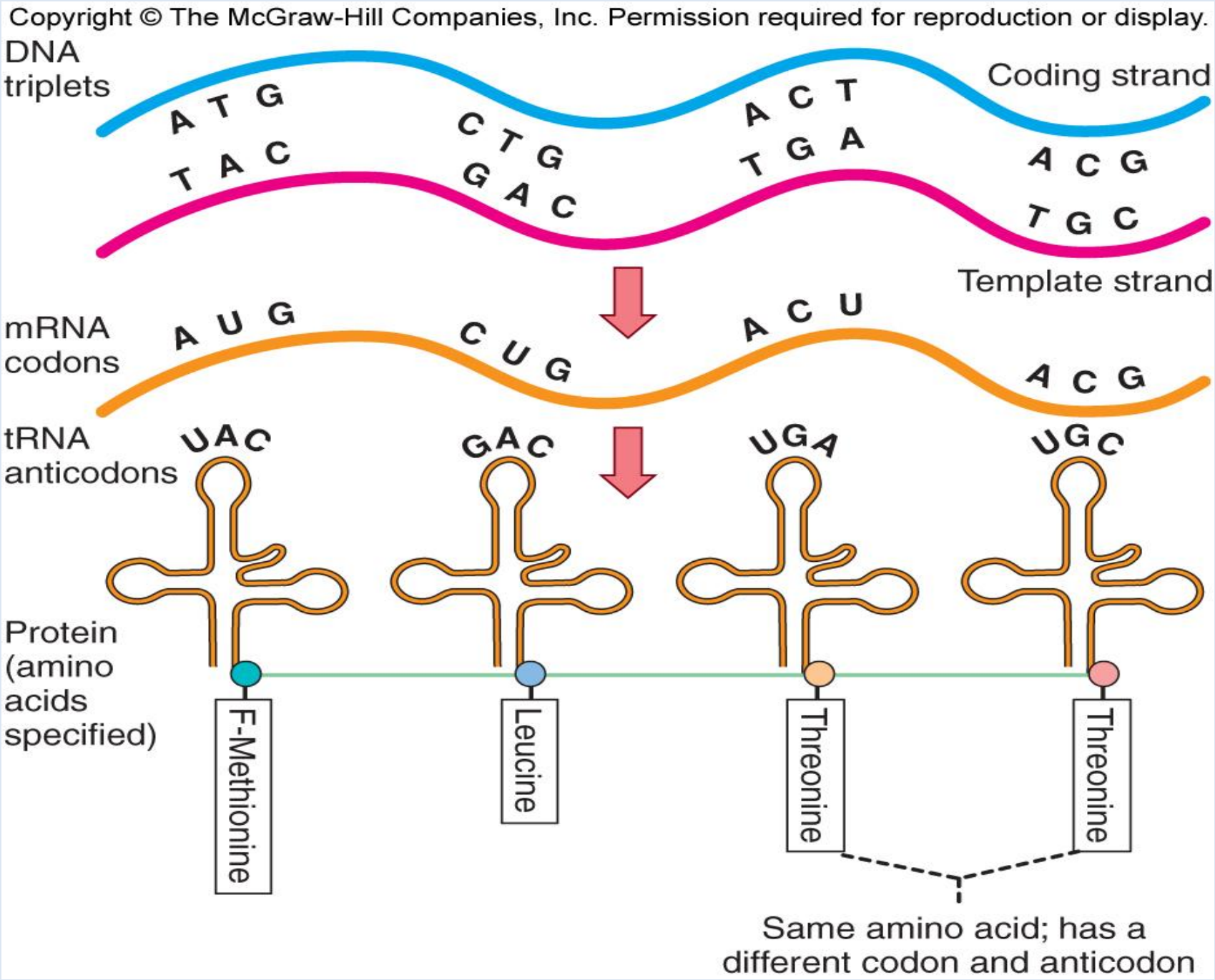
- The small subunit binds to the 5’ end of the mRNA
- Large subunit supplies enzymes for making peptide bonds on the protein
- The ribosome scans the mRNA by moving in the 5’ to 3’ direction along the mRNA
- The first codon is the START codon (AUG but can rarely be GUG)
- With the mRNA message in place on the ribosome, the tRNAs enter the ribosome with their amino acids
  - The complementary tRNA meets with the mRNA code
  - Guided by the two sites on the large subunit called the P site and the A site
  - The E site is where used tRNAs are released

The Master Genetic Code: The Message in Messenger RNA

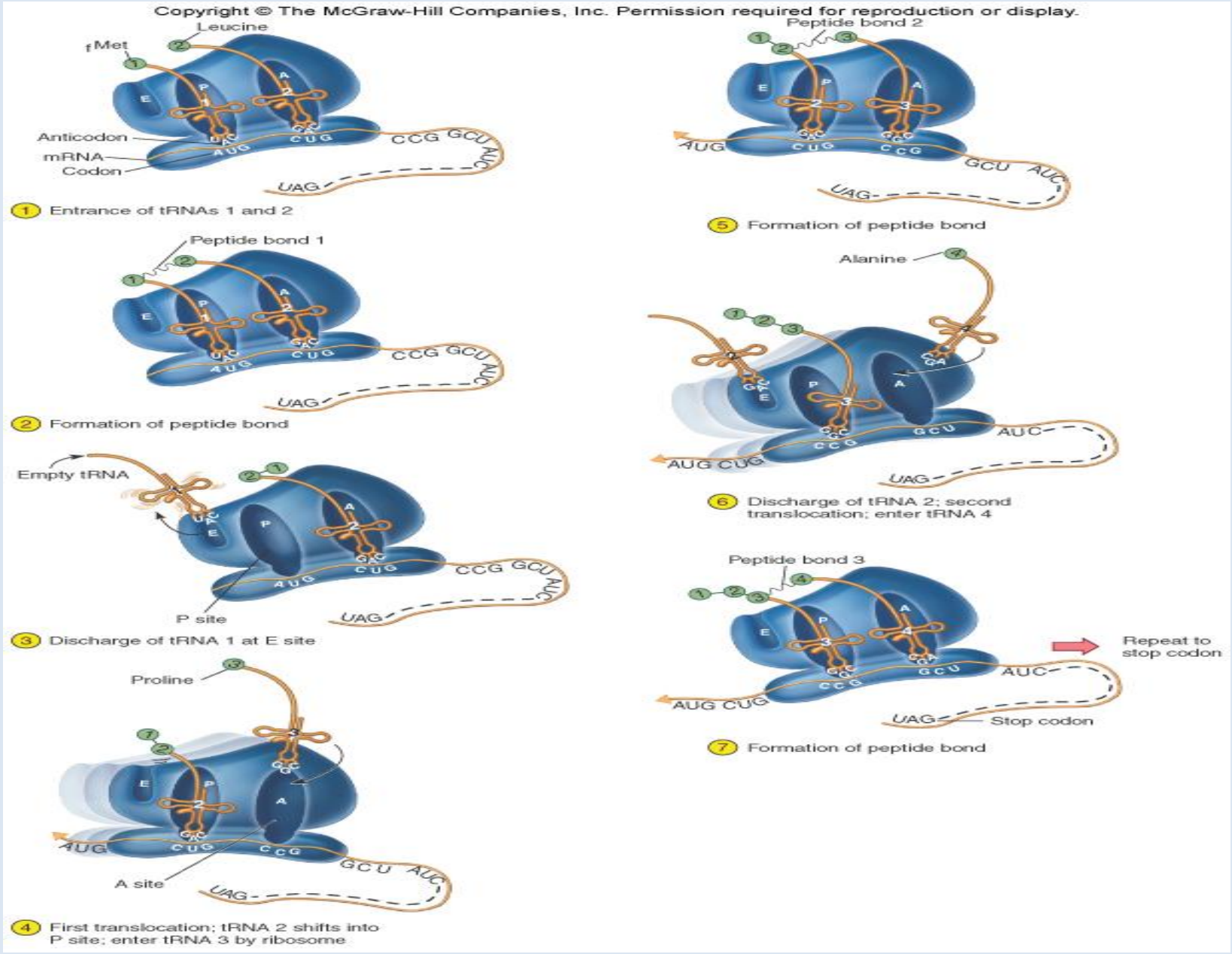
- The mRNA codons and the amino acids they specify
- **Redundancy** of the genetic code: a particular amino acid can be coded for by more than a single codon
- Wobble: in many cases, only the first two nucleotides are required to encode the correct amino acid- thought to permit some variation or mutation without altering the message

First Base Position	Second Base Position: U	Second Base Position: C	Second Base Position: A	Second Base Position: G	Third Base Position
U	UUU, UUC: Phenylalanine UUA, UUG: Leucine	UCU, UCC, UCA, UCG: Serine	UAU, UAC: Tyrosine UAA, UAG: STOP**	UGU, UGC: Cysteine UGA: STOP** UGG: Tryptophan	U, C, A, G
C	CUU, CUC, CUA, CUG: Leucine	CCU, CCC, CCA, CCG: Proline	CAU, CAC: Histidine CAA, CAG: Glutamine	CGU, CGC, CGA, CGG: Arginine	U, C, A, G
A	AUU, AUC: Isoleucine AUA: Isoleucine AUG: START; Methionine*	ACU, ACC, ACA, ACG: Threonine	AAU, AAC: Asparagine AAA, AAG: Lysine	AGU, AGC: Serine AGA, AGG: Arginine	U, C, A, G
G	GUU, GUC, GUA, GUG: Valine	GCU, GCC, GCA, GCG: Alanine	GAU, GAC: Aspartic acid GAA, GAG: Glutamic acid	GGU, GGC, GGA, GGG: Glycine	U, C, A, G

* This codon initiates translation.
**For these codons, which give the orders to stop translation, there are no corresponding tRNAs and no amino acids.



The Beginning of Protein Synthesis



Figure

The Termination of Protein Synthesis

- Brought about by the presence of a termination codon: UAA, UAG, and UGA

- Often called nonsense codons
- Do not code for a tRNA

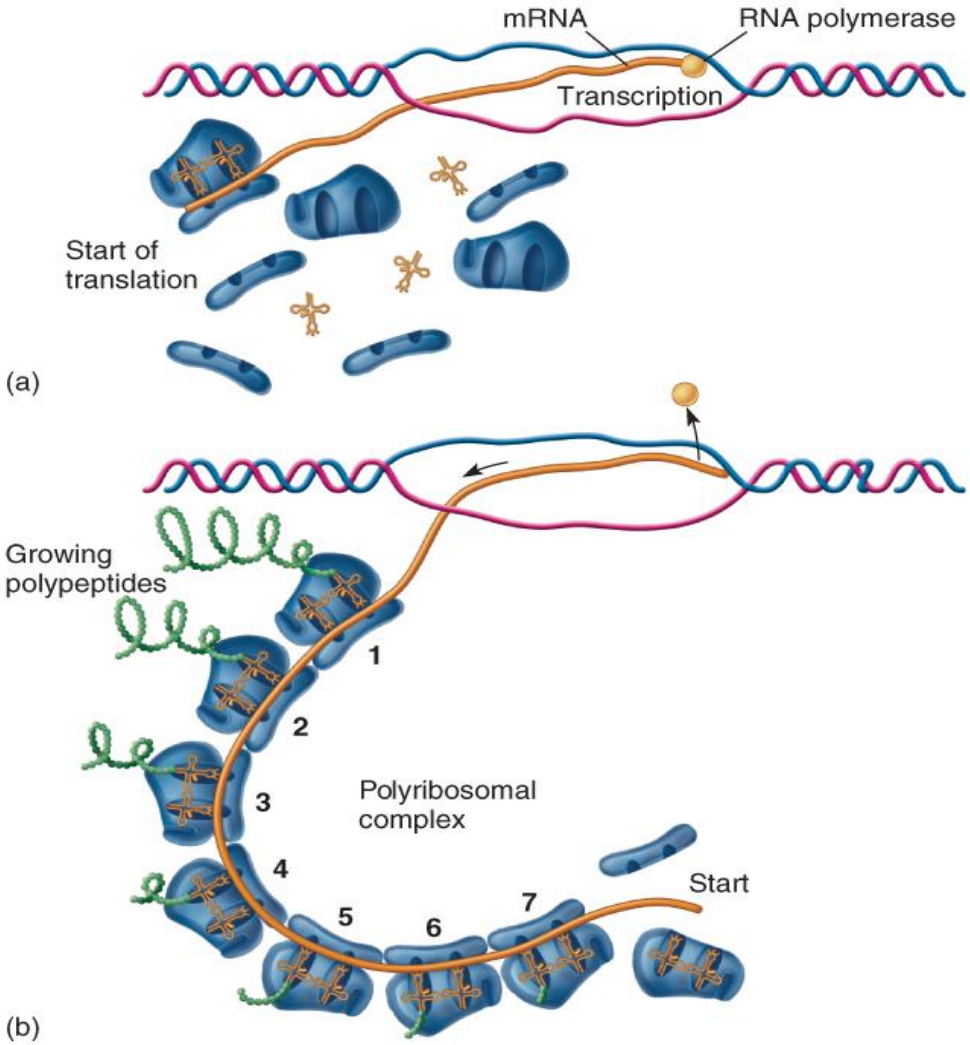
- When reached, a special enzyme breaks the bond between the final tRNA and the finished polypeptide chain, releasing the polypeptide chain from the ribosome

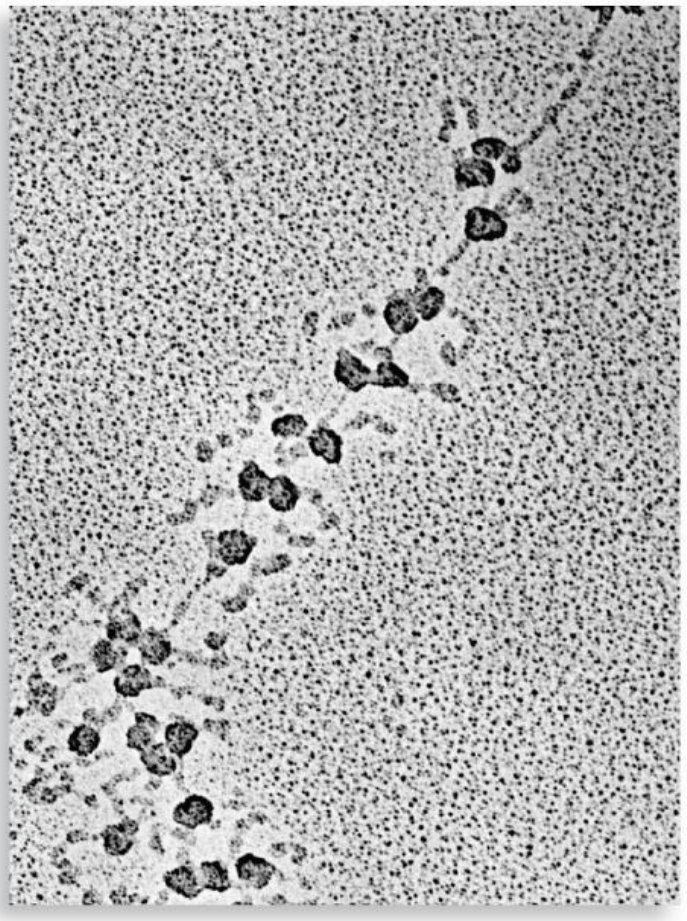
Modifications to Proteins

- Before it is released from the ribosome it starts to fold upon itself to achieve its biologically active tertiary conformation

- Post-translational modifications may be necessary
  - Starting animo acid (methionine) clipped off
  - Cofactors added
  - Join with other proteins to form quaternary levels of structure

Transcription and Translation is Efficient (Polyribosomes)





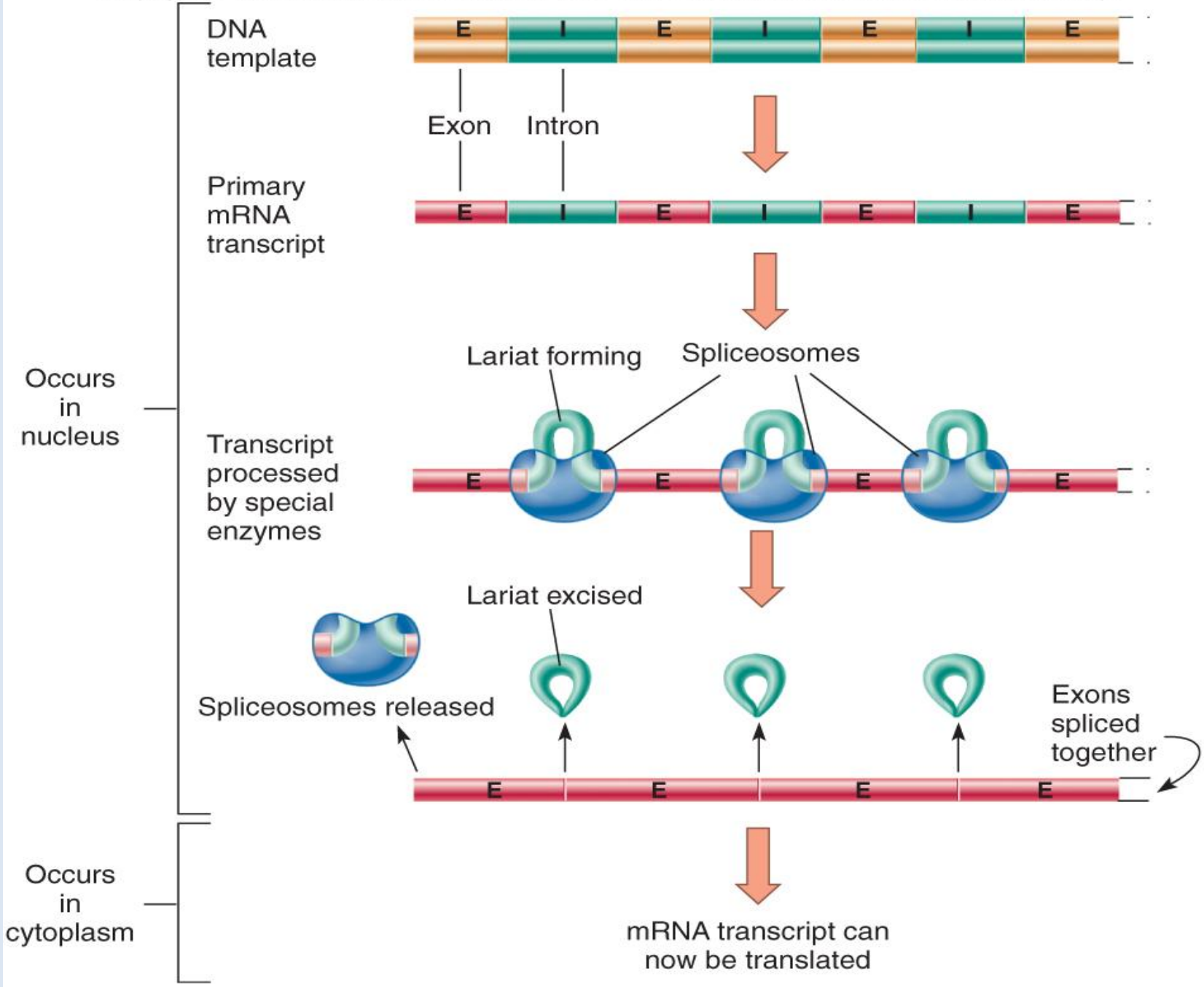
Eukaryotic Transcription and Translation: Similar Yet Different

- Start codon is also AUG, but it codes for a different form of methionine
- Eukaryotic mRNAs code for just one protein
- The presence of the DNA in the nucleus means that eukaryotic transcription and translation cannot be simultaneous
- mRNA in eukaryotes must pass through pores in the nuclear membrane and be carried to the ribosomes in the cytoplasm for translation

- Most eukaryotic genes do not exist as an uninterrupted series of triplets coding for a protein
  - **Introns**- sequences of bases that do not code for protein
  - **Exons**- coding regions that will be translated into protein
  - Called a split gene- requires further processing before translation
  - Transcription of the entire gene with both exons and introns occurs first, producing a pre-mRNA

Most eukaryotic genes do not exist as an uninterrupted series of triplets coding for a protein

- A series of adenosines is added to the mRNA molecule (protects it and directs it out of the nucleus)
- A splicesome recognizes the exon-intron junctions and enzymatically cuts through them
- The exons are joined end to end
- Some introns do code for cell substances (in humans, introns represent 98% of the DNA)



Figure

The Genetics of Animal Viruses

- Diverse
- Some- nucleic acid is linear; others, circular
- Most exist in a single molecule, but in a few it is in several
- Most contain dsDNA or ssRNA, but other patterns exist
- In all cases:
  - Viral nucleic acid penetrates the cell
  - The nucleic acid is introduced into the host's gene-processing machinery
  - The virus instructs the host's machinery to synthesize large numbers of new virus particles
  - Viral mRNA is translated into viral proteins on host cell ribosomes using host tRNA

9.3 Genetic Regulation of Protein Synthesis and Metabolism

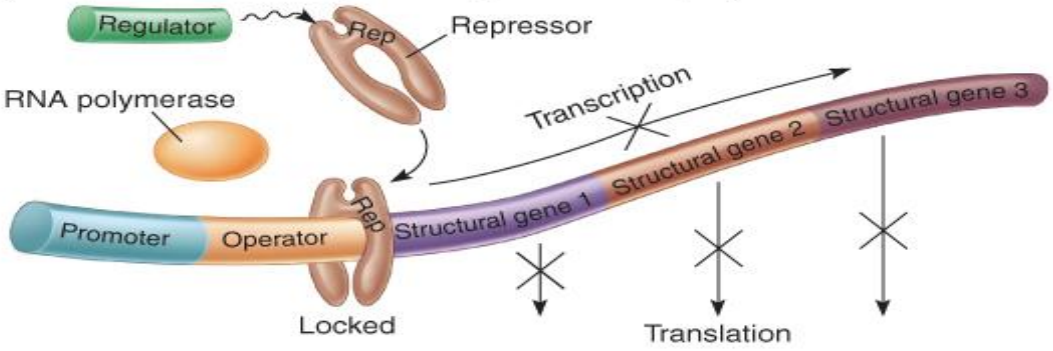
- Control mechanisms ensure that genes are active only when their products are required
  - Enzymes are produced as they are needed
  - Prevents the waste of energy and materials
  - Antisense RNAs, micro RNAs, and riboswitches provide regulation in prokaryotes and eukaryotes

- Prokaryotes organize collections of genes into **operons**
  - Coordinated set of genes regulated as a single unit
  - Either inducible or repressible
    - Inducible- the operon is turned in by the substrate of the enzyme for which the structural genes code
    - Repressible- contain genes coding for anabolic enzymes; several genes in a series are turned off by the product synthesized by the enzyme

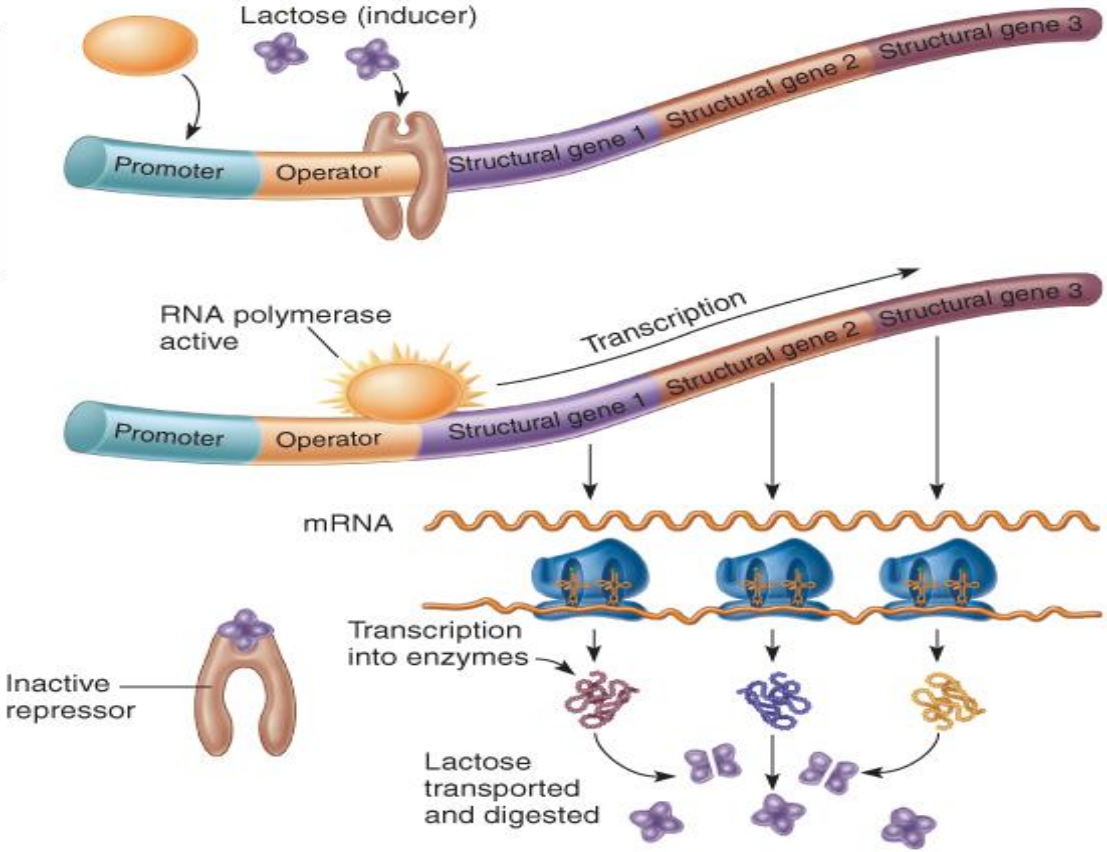
The Lactose Operon: A Model for Inducible Gene Regulation in Bacteria

- Best understood cell system for explaining control through genetic induction
- **Lactose (*lac*) operon**
- Regulates lactose metabolism in *Escherichia coli*
- Three important features:
  - The **regulator** (a gene that codes for a protein capable of repressing the operon [a **repressor**])
  - The control locus
    - **Promoter**- recognized by RNA polymerase
    - **Operator**- a sequence that acts as an on/off switch for transcription
  - The structural locus, made up of three genes each coding for a different enzyme needed to catabolize lactose

(a) **Operon Off.** In the absence of lactose, a repressor protein (the product of a regulatory gene located elsewhere on the bacterial chromosome) attaches to the operator of the operon. This effectively locks the operator and prevents any transcription of structural genes downstream (to its right). Suppression of transcription (and consequently, of translation) prevents the unnecessary synthesis of enzymes for processing lactose.



(b) **Operon On.** Upon entering the cell, the substrate (lactose) becomes a genetic inducer by attaching to the repressor, which loses its grip and falls away. The RNA polymerase is now free to bind to the promoter and initiate transcription, and the enzymes produced by translation of the mRNA perform the necessary reactions on their lactose substrate.



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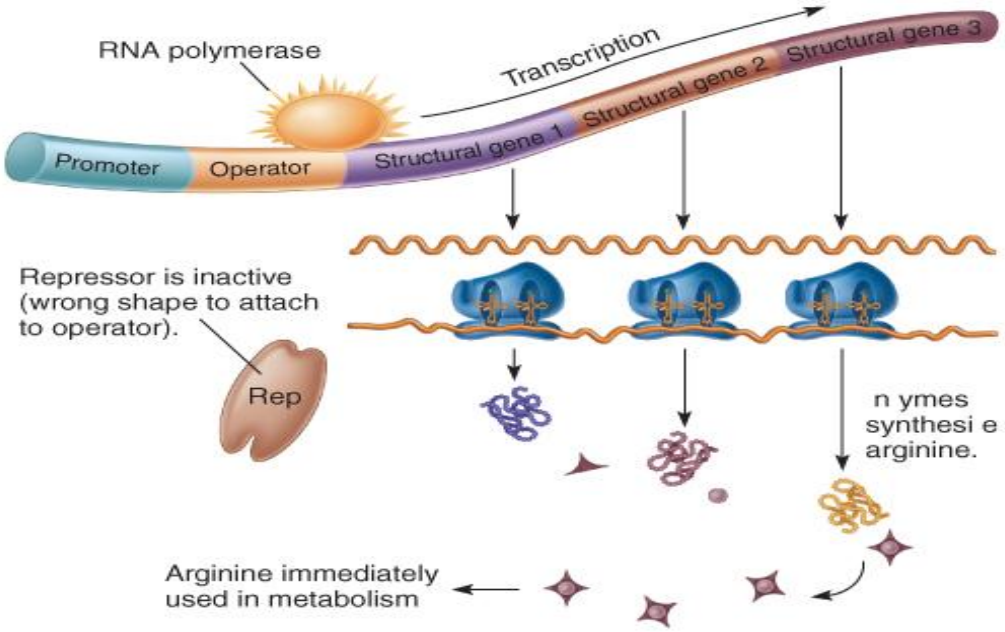
A Repressible Operon

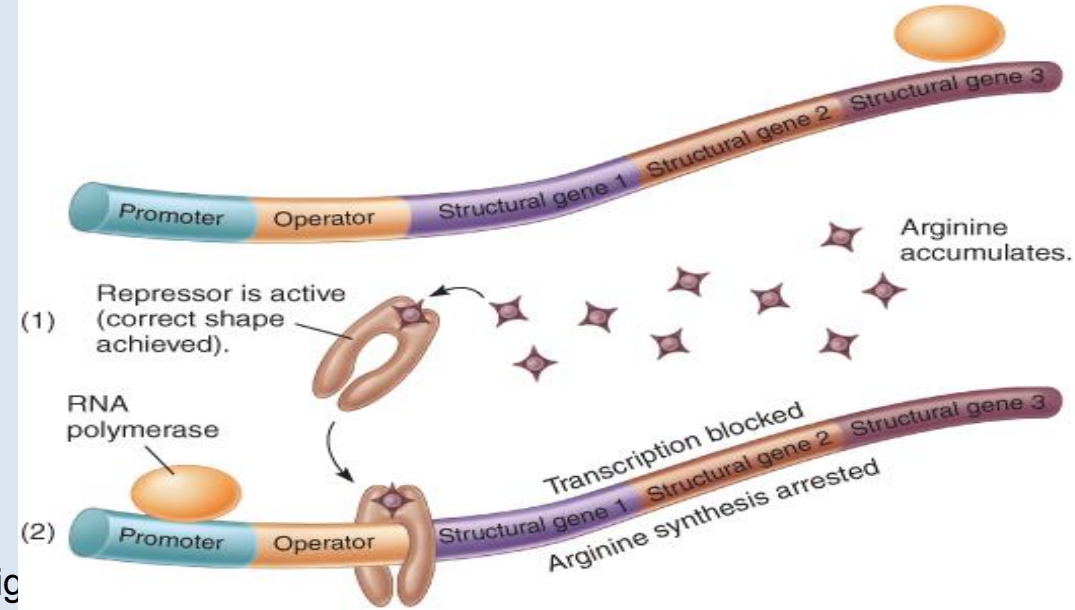
- Normally the operon is in the “on” mode and will be turned “off” only when the nutrient is no longer required

- The excess nutrient serves as a **corepressor** needed to block the action of the operon

- Example, *arg* operon



(a) **Operon On.** A repressible operon remains on when its nutrient products (here, arginine) are in great demand by the cell because the repressor is unable to bind to the operator at low nutrient levels.



(b) **Operon Off.** The operon is repressed when (1) arginine builds up and, serving as a corepressor, activates the repressor. (2) The repressor complex affixes to the operator and blocks the RNA polymerase and further transcription of genes for arginine synthesis.

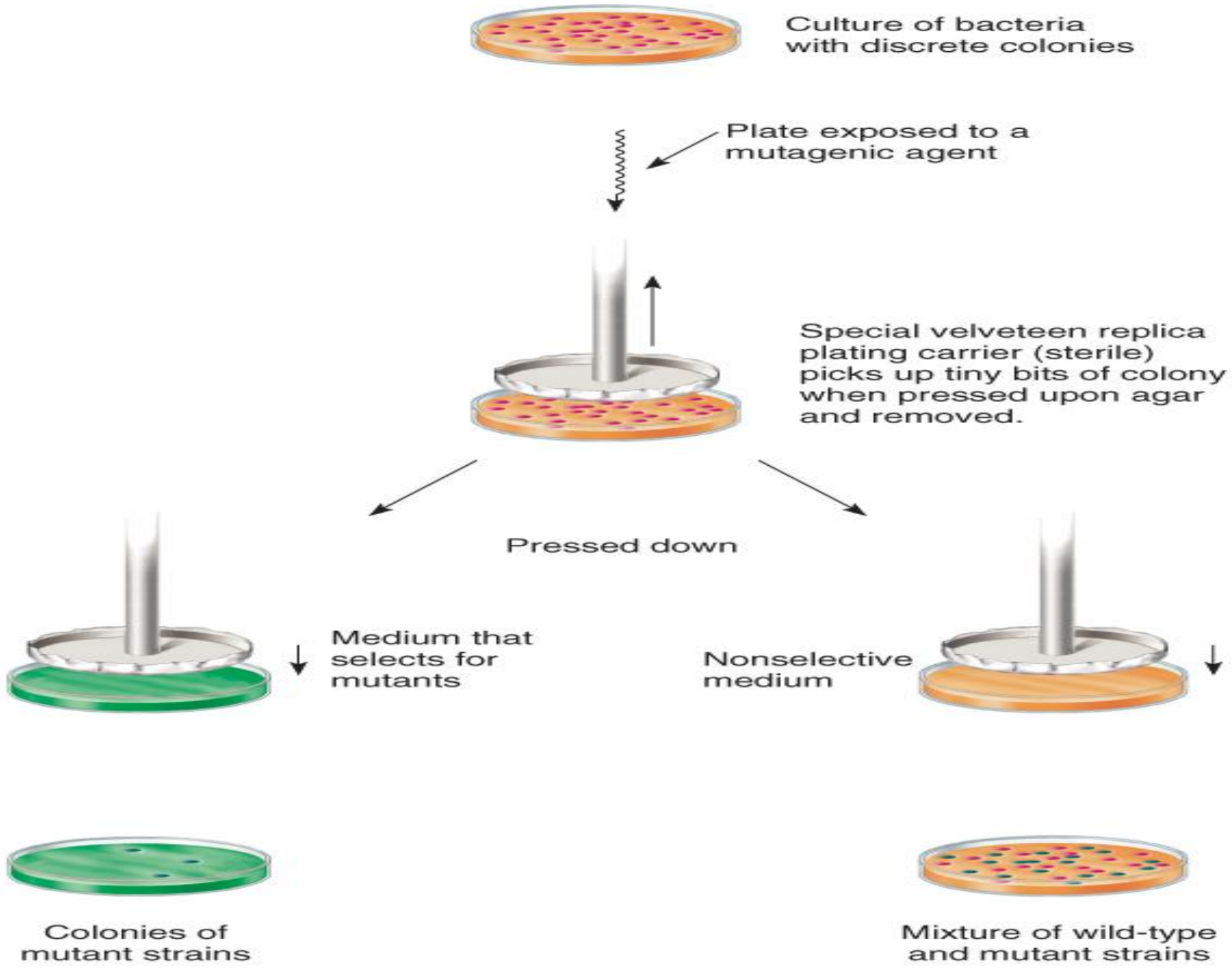
Fig

Antibiotics that Affect Transcription and Translation

- Some infection therapy is based on the concept that certain drugs react with DNA, RNA, or ribosomes and alter genetic expression
- Based on the premise that growth of the infectious agent will be inhibited by blocking its protein-synthesizing machinery selectively
- Drugs that inhibit protein synthesis exert their influence on transcription or translation
- Antibiotics often target the ribosome- inhibiting ribosomal function and ultimately protein synthesis

Mutations: Changes in the Genetic Code

- Genetic change is the driving force of evolution
- **Mutation**: when phenotypic changes are due to changes in the genotype
- An alteration in the nitrogen base sequence of DNA
- **Wild type**: a microorganism that exhibits a natural, nonmutated characteristic
- **Mutant strain**: when a microorganism bears a mutation
  - Useful for tracking genetic events,
  - Unraveling genetic organization, and
  - Pinpointing genetic markers



Causes of Mutations

- **Spontaneous mutation**: random change in the DNA arising from errors in replication

- **Induced mutation**: results from exposure to known mutagens

TABLE 9.3 Selected Mutagenic Agents and Their Effects

Agent	Effect
Chemical	
Nitrous acid, bisulfite	Removes an amino group from some bases
Ethidium bromide	Inserts between the paired bases
Acridine dyes	Cause frameshifts due to insertion between base pairs
Nitrogen base analogs	Compete with natural bases for sites on replicating DNA
Radiation	
Ionizing (gamma rays, X rays)	Form free radicals that cause single or double breaks in DNA
Ultraviolet	Causes cross-links between adjacent pyrimidines

Categories of Mutations

- **Point mutations**: involve addition, deletion, or substitution of single bases
  - **Missense mutation**: any change in the code that leads to placement of a different amino acid
    - Can create a faulty, nonfunctional protein
    - Can produce a protein that functions in a different manner
    - Can cause no significant alteration inl protein function
  - **Nonsense mutation**: changes a normal codon into a stop codon
  - **Silent mutation**: alters a base but does not change the amino acid and thus has no effect
  - **Back-mutation**: when a gene that has undergone mutation reverses to its original base composition

Categories of Mutations cont.

- **Frame shift mutations**:

  mutations that occur when one or more bases are inserted into or deleted from a newly synthesized DNA strand

  - Changes the reading frame of the mRNA
  - Nearly always result in a nonfunctional protein

TABLE 9.4 Classification of Major Types of Mutations

	Example
(a) Wild type (original, nonmutated sequence)	THE BIG BAD DOG ATE THE FAT RED CAT
Substitution mutations	
(b) Missense	THE BIG BAD DOG ATE THE FIT RED CAT
(c) Nonsense	THE BIG BAD (stop)
Frameshift mutations	
(d) Insertion	THE BIG BAB DDO GAT ETH EFA TRE DCA T
(e) Deletion	THE BIG BDD OGA TET HEF ATR EDC AT

Categories of mutations based on type of DNA alteration

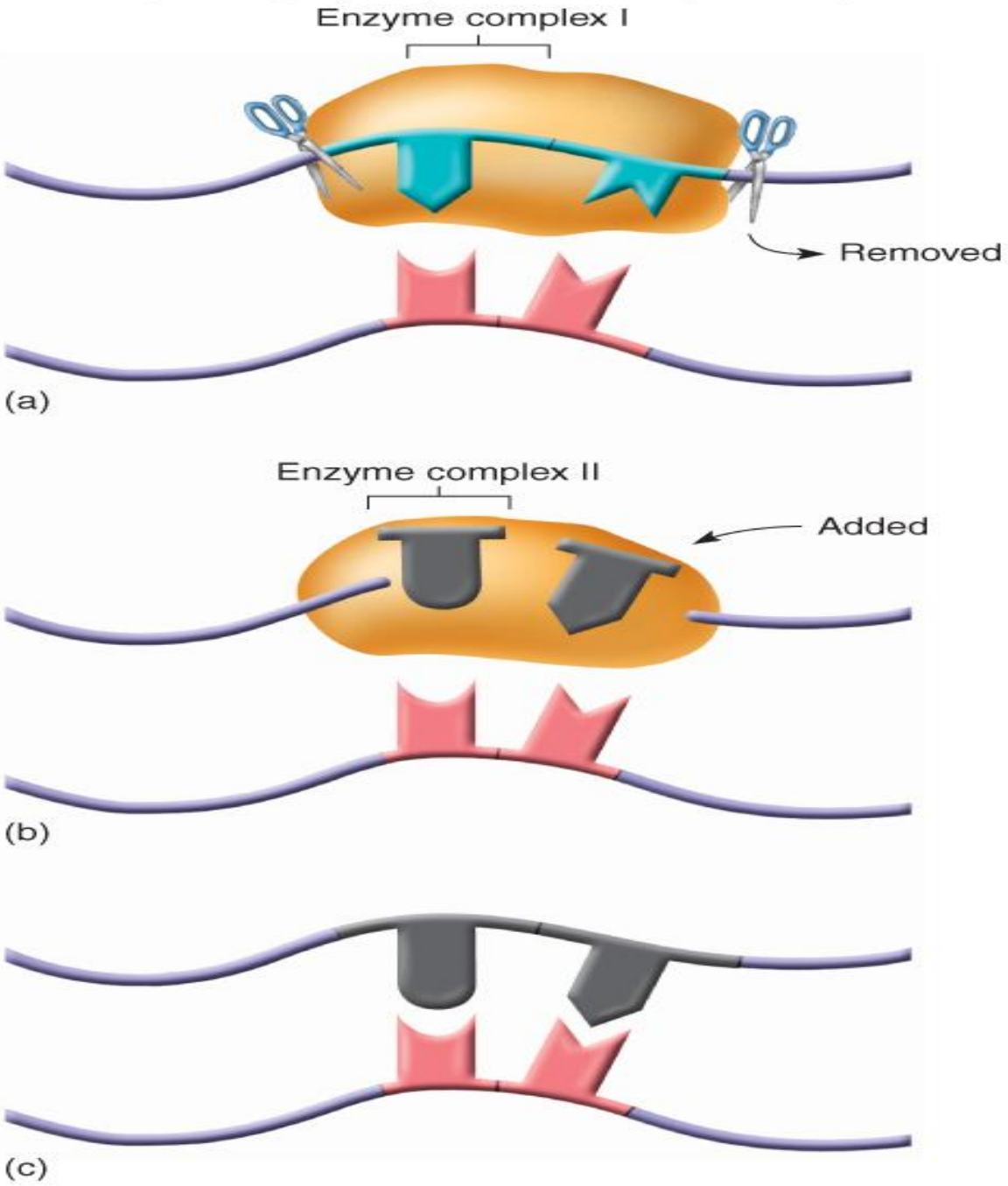
(a) The wild-type sequence of a gene is the DNA sequence found in most organisms and is generally considered the "normal" sequence. (b) A missense mutation causes a different amino acid to be incorporated into a protein. Effects range from unnoticeable to severe, based on how different the two amino acids are. (c) A nonsense mutation converts a codon to a stop codon, resulting in premature termination of protein synthesis. Effects of this type of mutation are almost always severe. (d, e) Insertion and deletion mutations cause a change in the reading frame of the mRNA, resulting in a protein in which every amino acid after the mutation is affected. Because of this, frameshift mutations almost always result in a nonfunctional protein.

Repair of Mutations

- Most ordinary DNA damage is resolved by enzymatic systems specialized for finding and fixing such defects

- DNA that has been damaged by UV radiation
  - Restored by photoactivation or light repair
  - DNA photolayse- light-sensitive enzyme

- Excision repair
  - Excise mutations by a series of enzymes
  - Remove incorrect bases and add correct one



The Ames Test

- Rapid screening system
- Detects chemicals with carcinogenic potential
- Any chemical capable of mutating bacterial DNA can similarly mutate mammalian DNA

Culture of *Salmonella* bacteria, histidine(⁻)

In the control setup, bacteria are plated on a histidine-free medium containing liver enzymes but lacking the test agent.

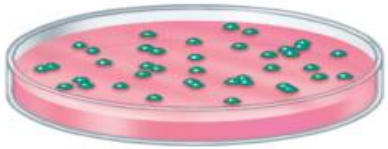
(a) Control Plate
Minimal medium with no histidine and no test chemical

(b) Test Plate
Minimal medium with test chemical and no histidine

The experimental plate is prepared the same way except that it contains the test agent. After incubation, plates are observed for colonies. Any colonies developing on the plates are due to a back-mutation in a cell, which has reverted it to a histidine(⁺) strain.

Incubation (12 h)
Any colonies that form have back-mutated to histidine(⁺).

Histidine(⁺) colonies arising from spontaneous back-mutation



Histidine(⁺) colonies induced by the chemical

(c) The degree of mutagenicity of the chemical agent can be calculated by comparing the number of colonies growing on the control plate with the number on the test plate. Chemicals that produce an increased incidence of back-mutation are considered carcinogens.

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Positive and Negative Effects of Mutations

- Mutations are permanent and inheritable
- Most are harmful but some provide adaptive advantages

DNA Recombination Events

- **Recombination**: when one organism donates DNA to another organism

- The end result is a new strain different from both the donor and the original recipient

- Bacterial plasmids and gene exchange

- **Recombinant** organism: Any organism that contains (and expresses) genes that originated in another organism

Transmission of Genetic Material in Bacteria

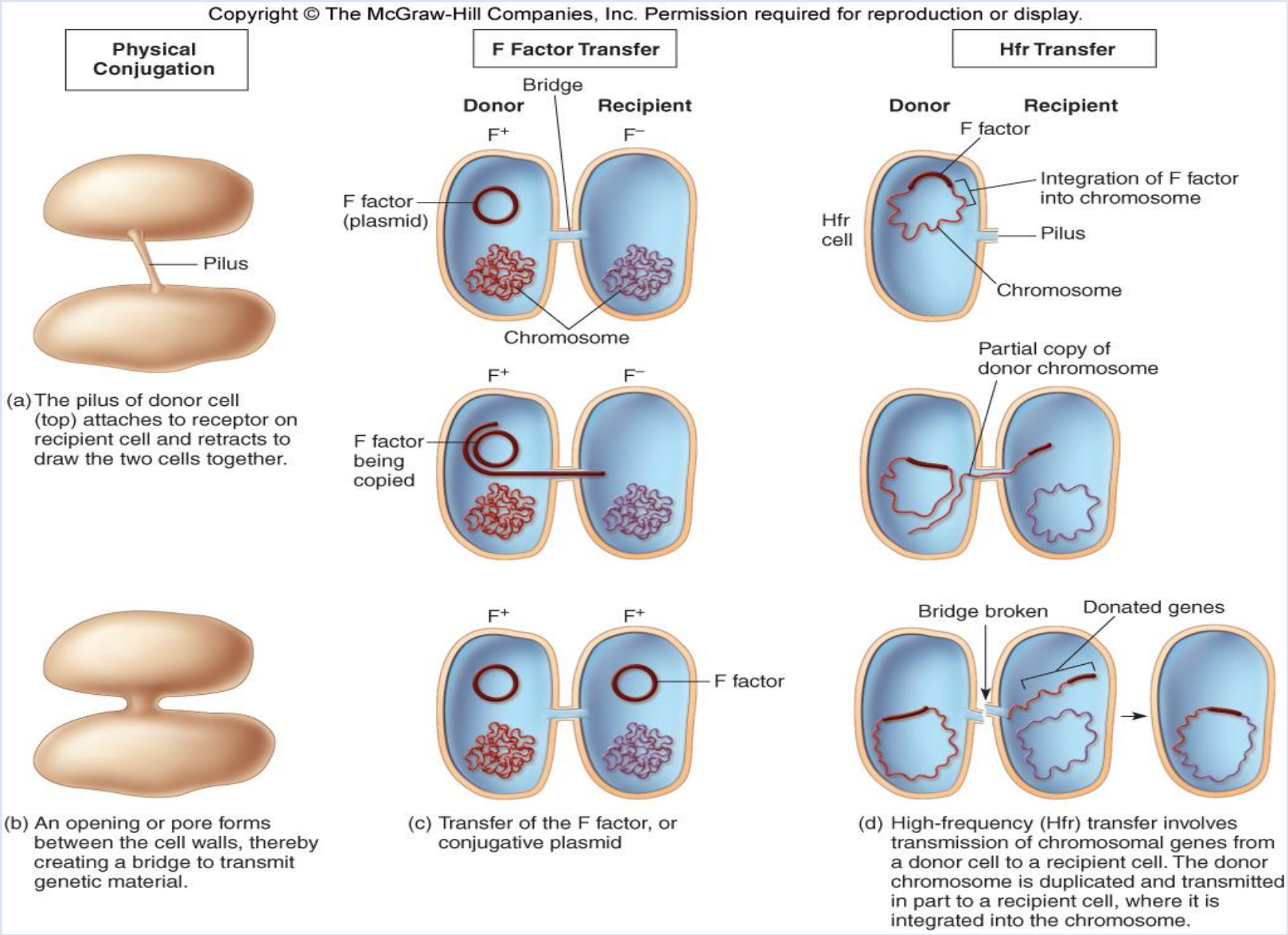
- Usually involves small pieces of DNA (plasmids or chromosomal fragments)
- Plasmids can replicate independently of the bacterial chromosome
- Chromosomal fragments must integrate themselves into the bacterial chromosome in order to replicate
- Three means of genetic recombination in bacteria
  - **Conjugation**
  - **Transformation**
  - **Transduction**

TABLE 9.5 Types of Intermicrobial Exchange

Mode	Factors Involved	Direct or Indirect*	Examples of Genes Transferred
Conjugation	Donor cell with pilus Fertility plasmid in donor Both donor and recipient alive Bridge forms between cells to transfer DNA.	Direct	Drug resistance; resistance to metals; toxin production; enzymes; adherence molecules; degradation of toxic substances; uptake of iron
Transformation	Free donor DNA (fragment) Live, competent recipient cell	Indirect	Polysaccharide capsule; unlimited with cloning techniques
Transduction	Donor is lysed bacterial cell. Defective bacteriophage is carrier of donor DNA. Live recipient cell of same species as donor	Indirect	Toxins; enzymes for sugar fermentation; drug resistance

*Direct means the donor and recipient are in contact during exchange; indirect means they are not.

Conjugation: Bacterial "Sex"



(a) The pilus of donor cell (top) attaches to receptor on recipient cell and retracts to draw the two cells together.

(b) An opening or pore forms between the cell walls, thereby creating a bridge to transmit genetic material.

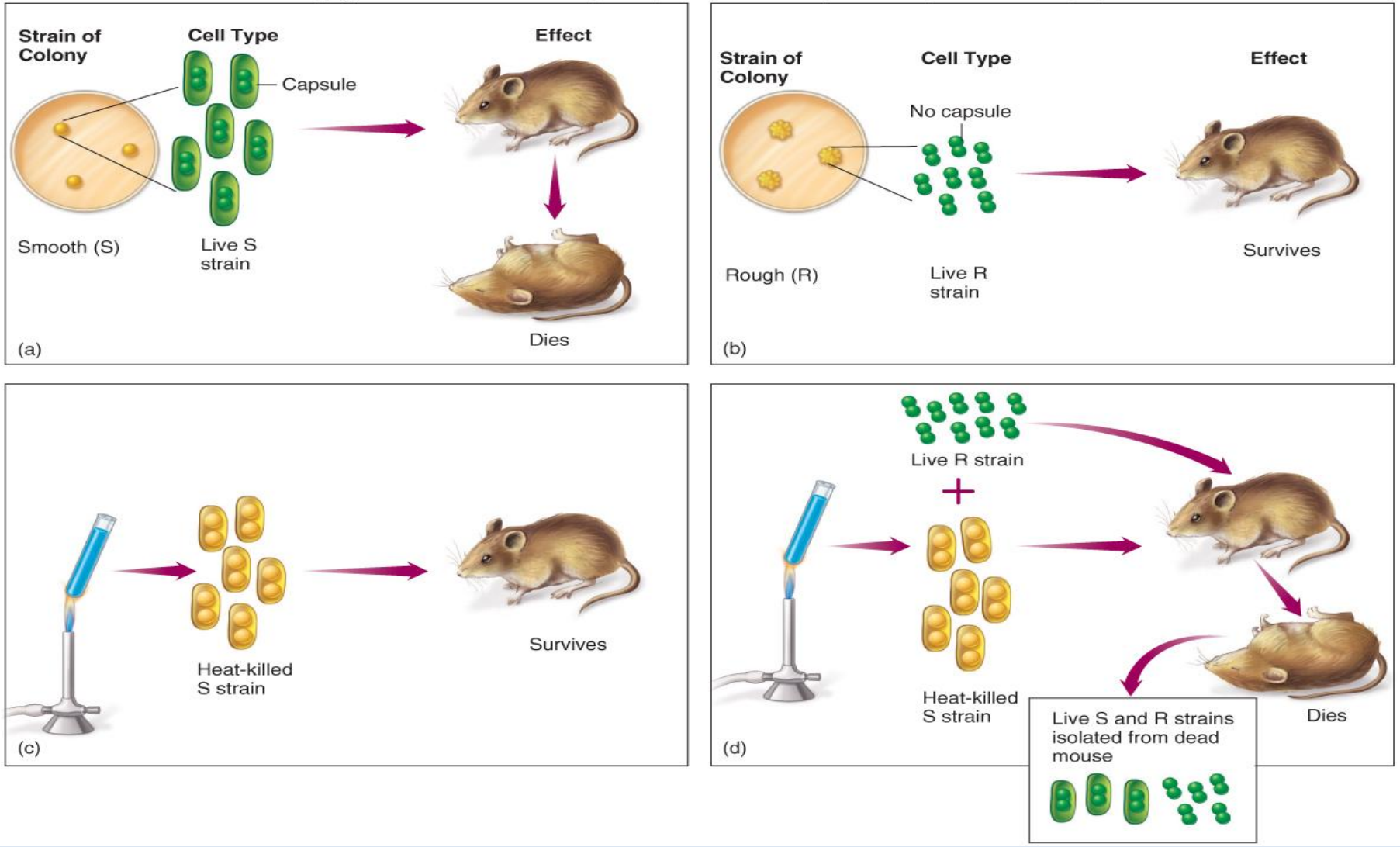
(c) Transfer of the F factor, or conjugative plasmid

(d) High-frequency (Hfr) transfer involves transmission of chromosomal genes from a donor cell to a recipient cell. The donor chromosome is duplicated and transmitted in part to a recipient cell, where it is integrated into the chromosome.

Biomedical Importance of Conjugation

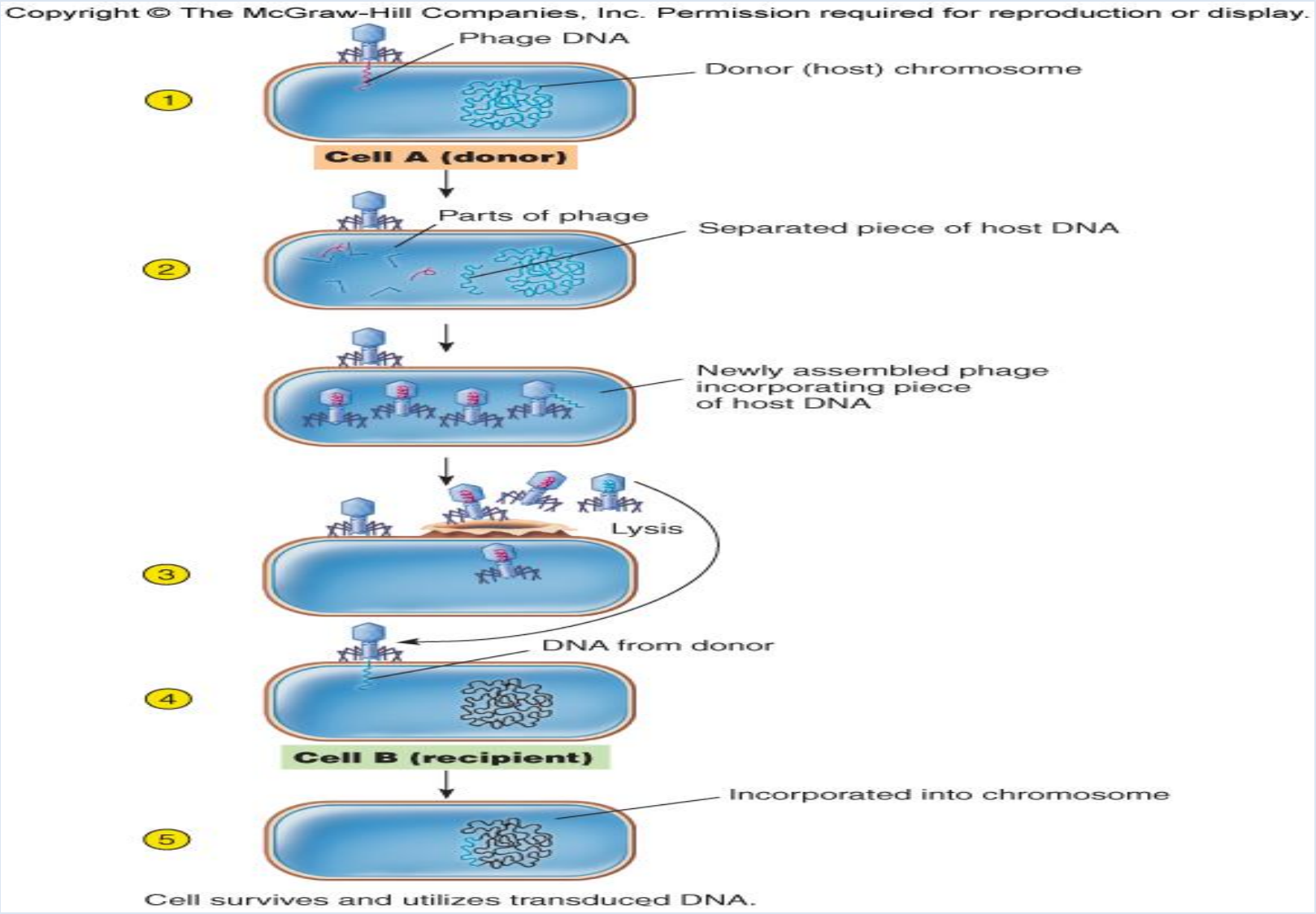
- **Resistance (R) plasmids**, or **factors**- bear genes for resisting antibiotics
- Can confer multiple resistance to antibiotics to a strain of bacteria
- R factors can also carry resistance to heavy metals or for synthesizing virulence factors

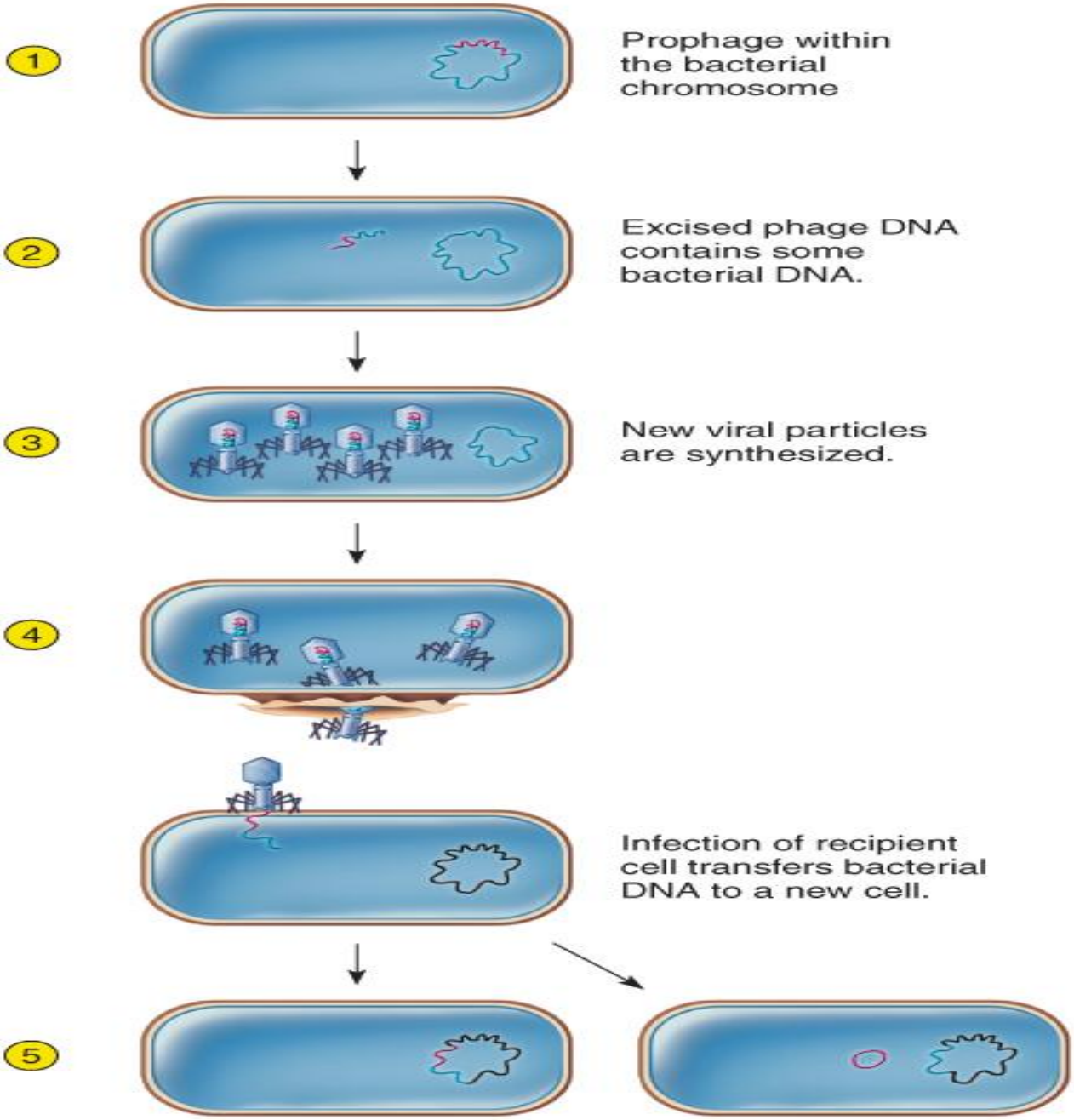
Transformation: Capturing DNA from Solution



- Griffith demonstrated that DNA released from a killed cell can be acquired by a live cell
  - Later studies supported this
  - Nonspecific acceptance by a bacterial cell- transformation
  - Facilitated by special DNA-binding proteins on the cell wall
  - Competent cells- capable of accepting genetic material
  - Useful for certain types of recombinant DNA technology

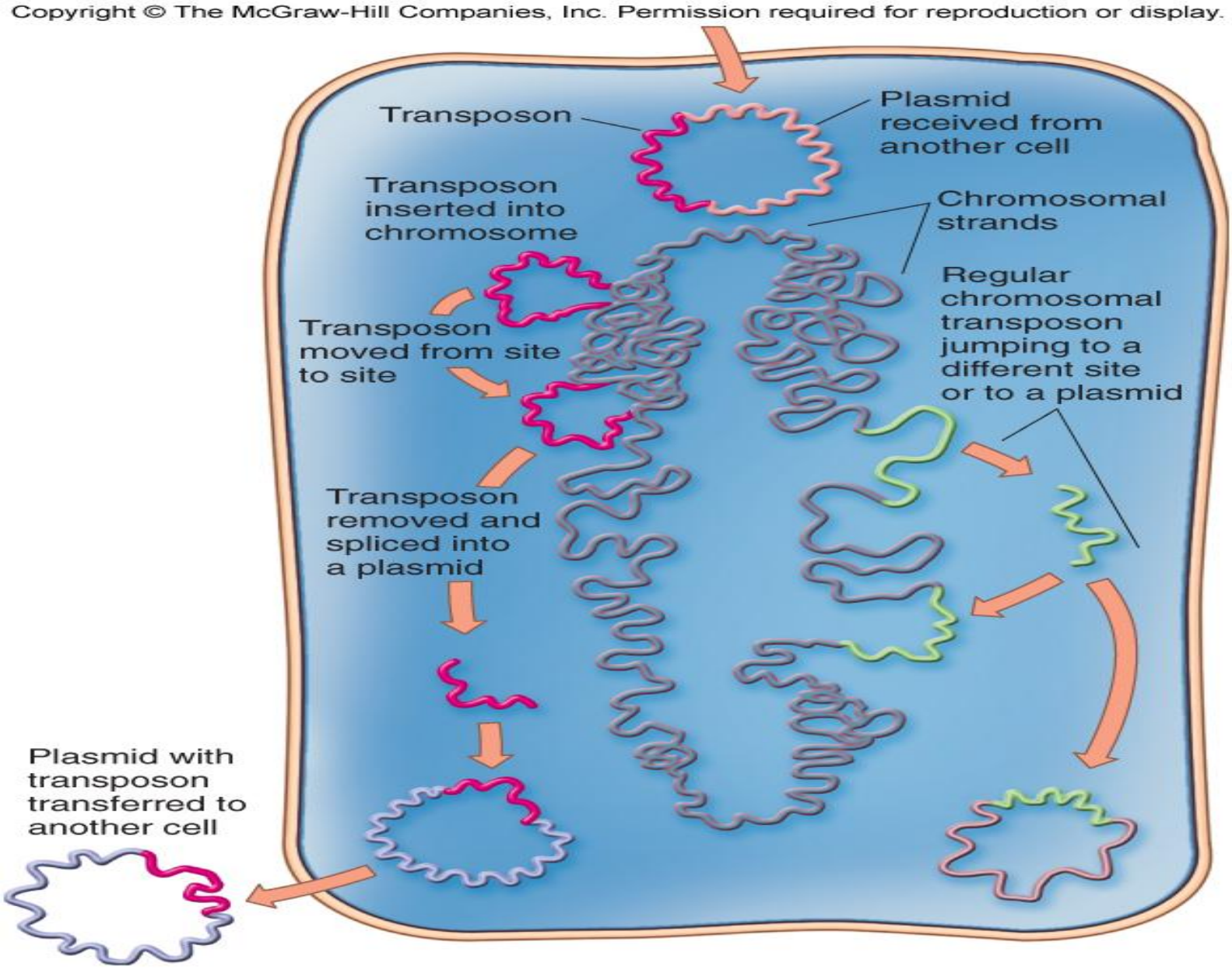
Transduction: The Case of the Piggyback DNA





Recombination results in two possible outcomes.

Transposons: “This Gene is Jumpin”



- Contain DNA that codes for the enzymes needed to remove and reintegrate the transposon at another site in the genome

- Insertion elements- tranposons that consist of only two genetic sequences

- Retro-transposon- can transcribe DNA into RNA and back into DNA for insertion in a new location

- Overall effect- scrambles the genetic language

- In bacteria, involved in:
  - Changes in traits such as colony morphology, pigmentation, and antigenic characteristics
  - Replacement of damaged DNA,
  - Inter-microbrial transfer of drug resistance