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Bacterial Genome Replication and Expression

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Terminology and Concepts

- **Genome**
  - all DNA present in a cell or virus
    - *Bacteria* and *Archaea* generally have one set (haploid – 1N)
    - eukaryotes have two sets (diploid - 2N)
- **Genotype**
  - specific set of genes an organism possesses
- **Phenotype**
  - collection of observable characteristics

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DNA and RNA Structure

- The nucleic acids**, DNA and RNA, are polymers of nucleotides**
  - linked together by **phosphodiester bonds**
- DNA and RNA differ in
  - **the nitrogenous bases they contain**
  - **the sugars they contain**
  - **whether they are single or double stranded**

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DNA Structure

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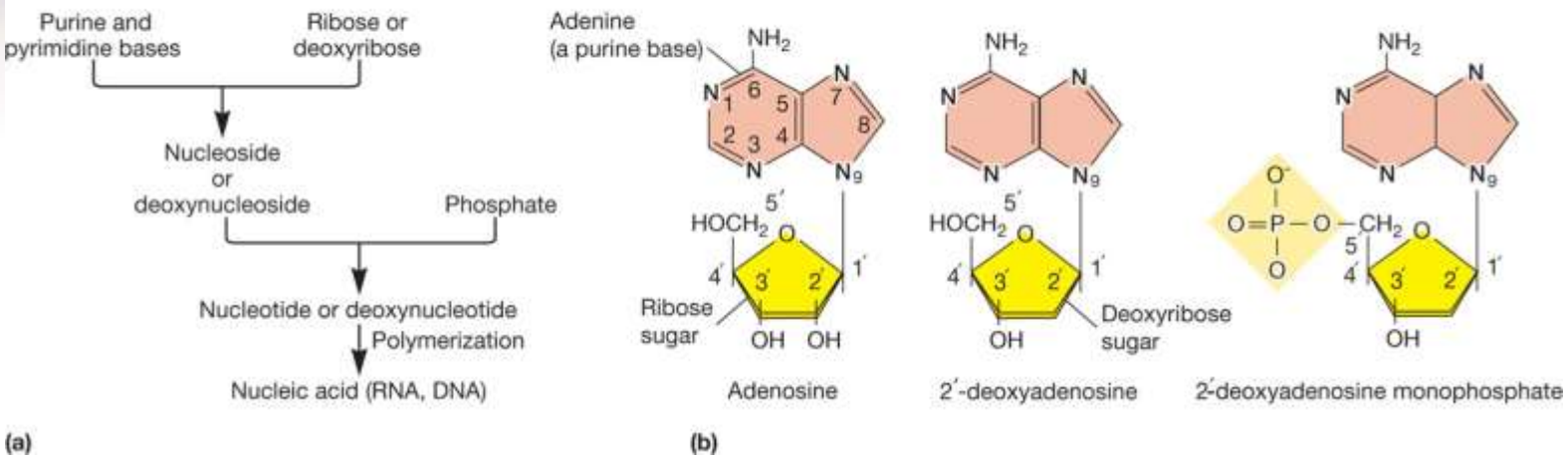

- Polymer of nucleotides
  - bases are adenine, guanine, cytosine, and thymine
  - sugar is deoxyribose
  - phosphate is esterified(مُجَمع) to sugar carbon
- Sugar phosphate backbone
  - covalent bonds between the 3’-hydroxyl of one sugar and a 5’-phosphate attached to an adjacent sugar

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DNA Structure – Two Complementary Strands

- Molecule is double stranded helix
- **Base pairing**
  - **adenine (purine) and thymine (pyrimidine) pair by 2 hydrogen bonds**
  - **guanine (purine) and cytosine (pyrimidine) pair by 3 hydrogen bonds**
- Major and minor grooves form when the 2 strands twist around each other

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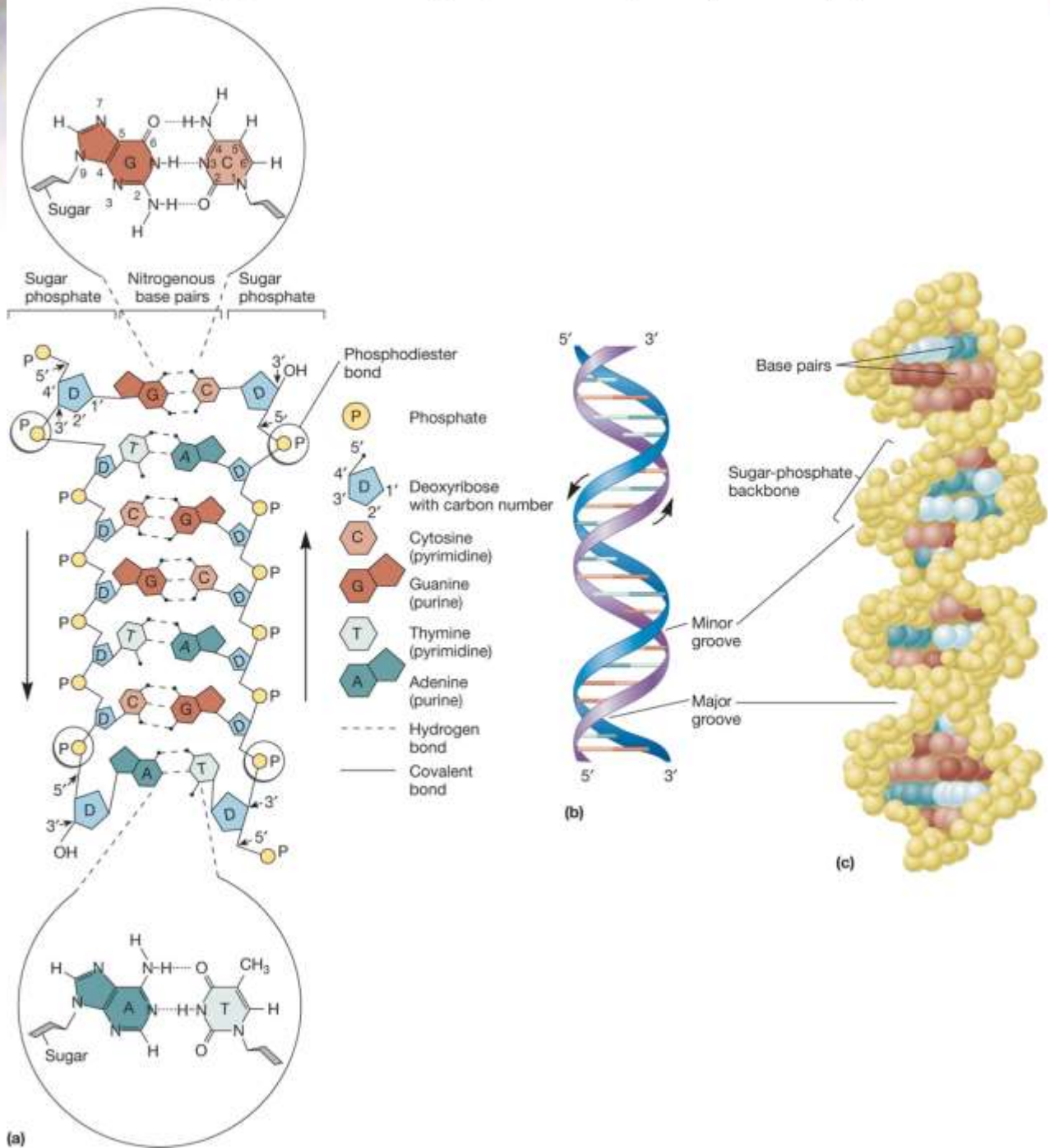

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RNA Structure

- Polymer of nucleotides
  - **contains the bases adenine, guanine, cytosine, and uracil**
  - **sugar is ribose**
  - **phosphodiester bonds**
- **Most RNA molecules are single stranded; some are double stranded**

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RNA Structure

- Three different types which may differ from each other in function, site of synthesis, and in structure
  - **messenger RNA (mRNA)**
  - **ribosomal RNA (rRNA)**
  - **transfer RNA (tRNA)**

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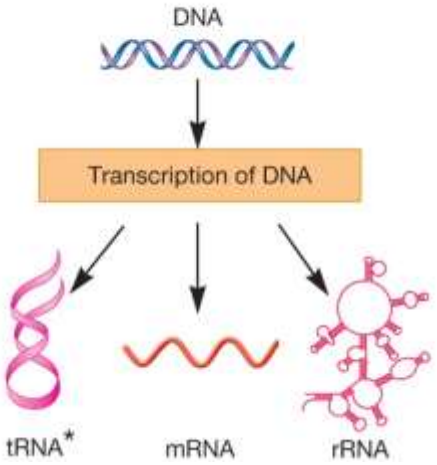

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Protein Structure

- Polymers of amino acids linked by peptide bonds
  - amino acids have central carbon
    - carboxy group (**C-terminal**)
    - amino group **(N-terminal**)
    - side chain
  - amino acids can be polar, non-polar, or charged depending on side chain

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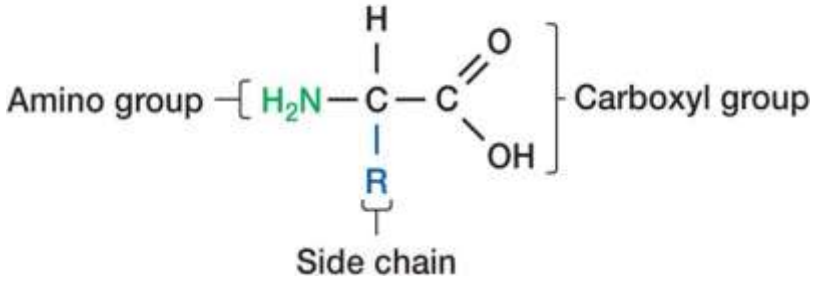

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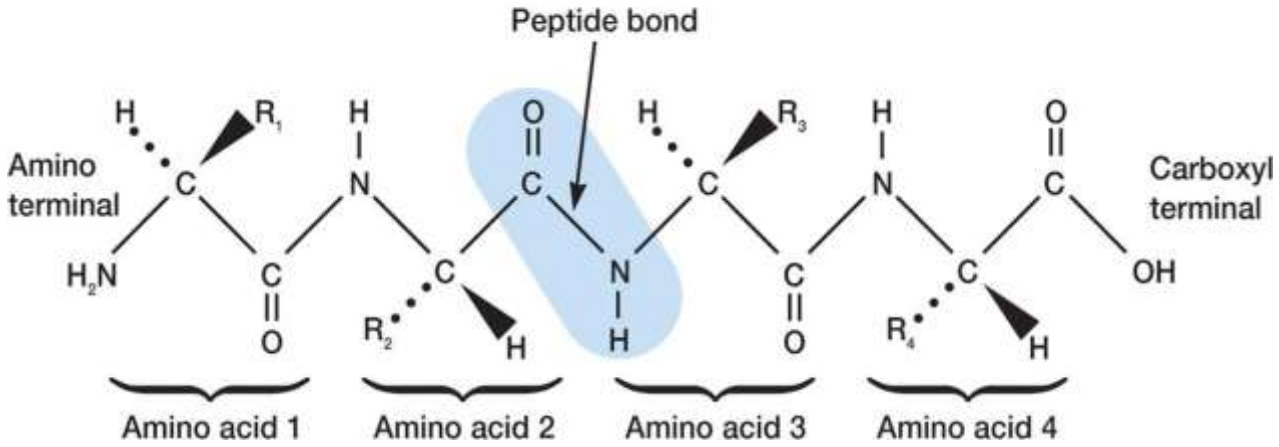

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Gene Structure

- Gene
  - **the basic unit of genetic information**
  - **also defined as the nucleic acid sequence that codes for a polypeptide, tRNA or rRNA**
  - **linear sequence of nucleotides with a fixed start point and end point**
  - **codons are found in mRNA and code for single amino acids**

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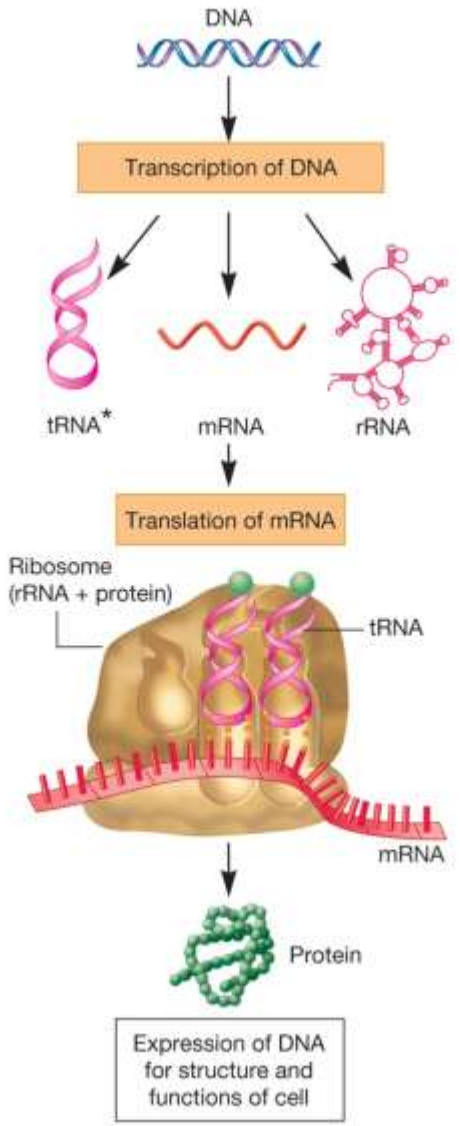

*The sizes of RNA are enlarged to show details.

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Protein-Coding Genes - 1

- Template strand of DNA directs RNA synthesis
  - is read in the 3’ to 5’ direction
- Complementary DNA strand
  - is coding strand, same nucleotide sequence as mRNA (except in thymine)

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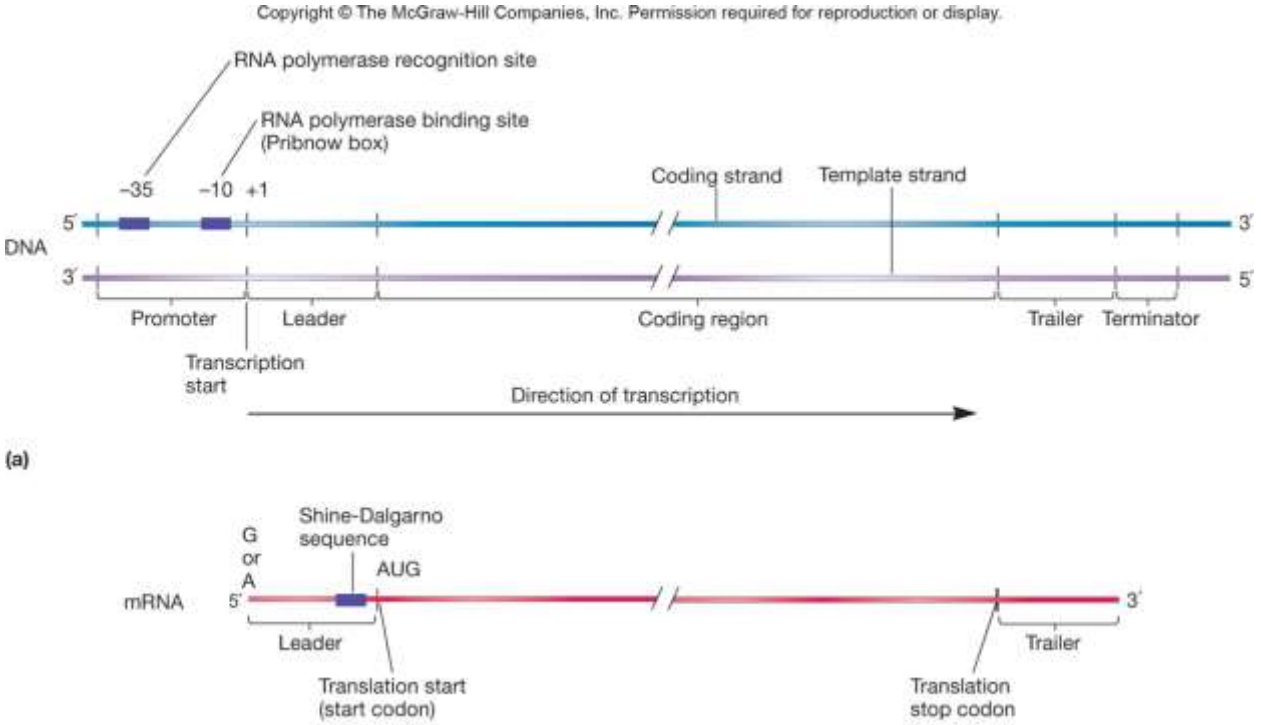

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Protein-Coding Genes - 2

- Promoter is located at the start of the gene
  - **is the recognition/binding site for RNA polymerase**
  - **functions to orient polymerase**
- **Leader sequence** is **transcribed into mRNA but is not translated into amino acids**
  - **Shine-Dalgarno sequence important for initiation of translation**

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Protein-Coding Genes - 3

- Begins with DNA sequence 3'-TAC-5'
  - produces codon AUG
  - codes for **N-formylmethionine**, a **modified amino acid used to initiate protein synthesis in bacteria**
  - coding region ends with **a stop codon**
    - immediately followed by the **trailer sequence which contains a terminator sequence used to stop transcription**

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tRNA and rRNA Genes

- DNA sequences that code for tRNA and rRNA are considered genes
  - genes coding for **tRNA may code for more than a single tRNA molecule or type of tRNA**
  - genes coding for rRNA are transcribed as single, large precursor
  - Spacers(الفواصل) between the coding regions of both are removed after transcription, some by the use of special ribonucleases called **ribozymes**

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Protein Maturation and Secretion

- Protein function depends on 3-D shape
- Occurs as post translational event
  - **requires folding**
  - **association with other proteins**
  - **delivered to proper subcellular or extracellular site**

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Protein Folding and Molecular Chaperones

- **Molecular chaperones**
  - proteins that aid the folding of nascent polypeptides
  - protect cells from thermal damage
    - **e.g., heat-shock proteins**
  - aid in transport of proteins across membranes

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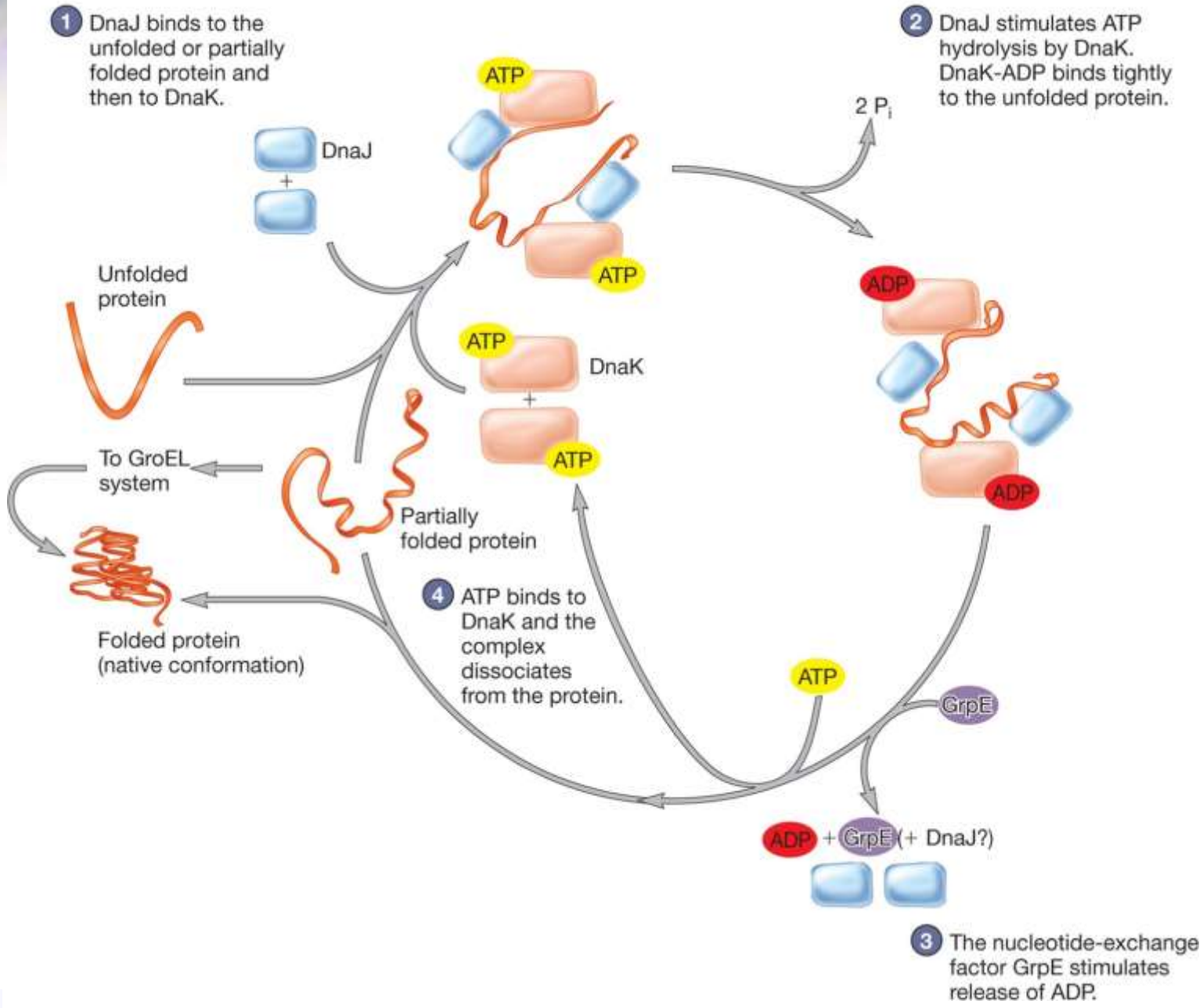

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Protein Splicing

- **Removal of part of polypeptide before folding**
- **Inteins – removed portion**
- **Exteins – portions that remain in protein**

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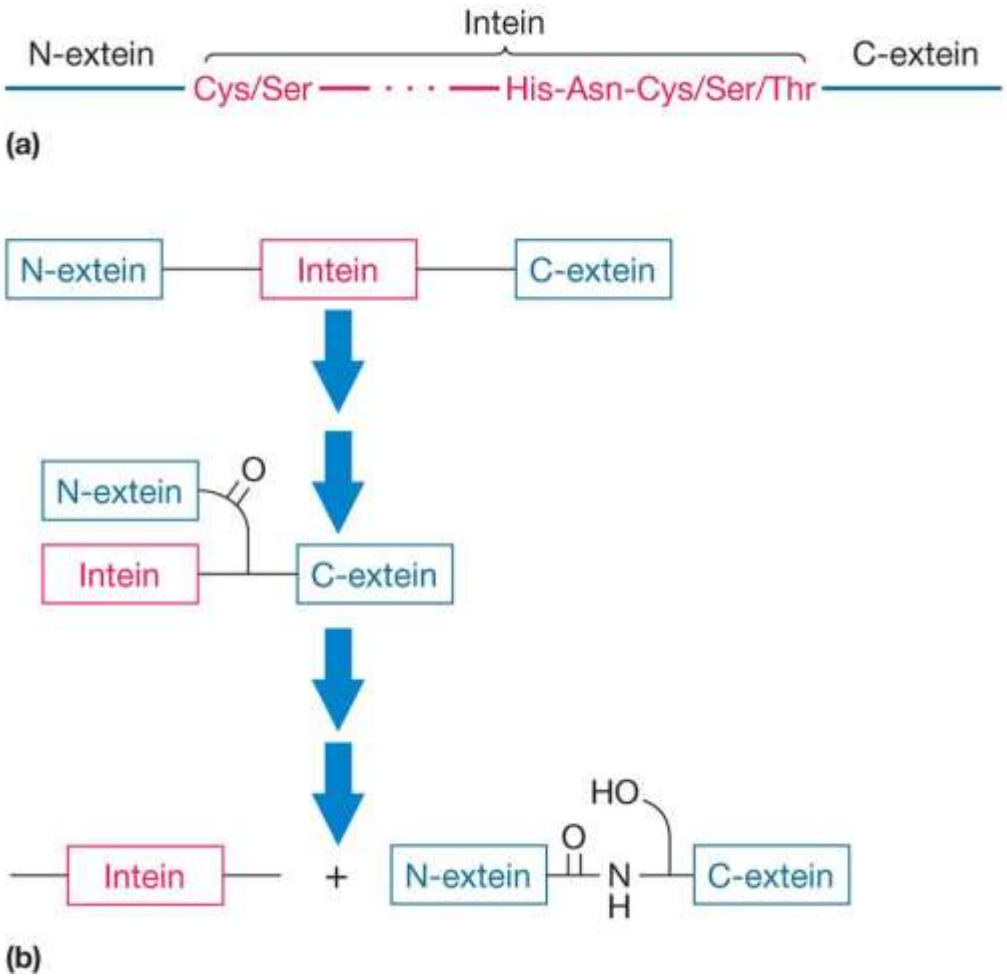

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Protein Translocation and Secretion in *Bacteria* - 1

- Numerous protein secretion pathways have been identified
  - **some reside(يُقيم) in all 3 domains**
  - **some unique to *Bacteria* and *Archaea***
  - **some unique to gram-negative cells**

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Protein Translocation and Secretion in *Bacteria* - 2

- **Translocation**
  - movement **of proteins from cytoplasm to plasma membrane or periplasmic space**
    - include transport proteins, ETC proteins, proteins involved in chemotaxis and cell wall synthesis, enzymes
- **Secretion**
  - movement of proteins from the cytoplasm to external environment
    - hydrolytic enzymes for nutrient break down

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Common Translocation and Secretion Systems

- **Sec-dependent pathway**
  - the major pathway for all bacteria for transporting proteins across the plasma membrane
- **Gram-negative bacteria**
  - may use Sec system
  - **also must cross the outer membrane using Types I, II, III, IV, V systems**
- All pathways require energy

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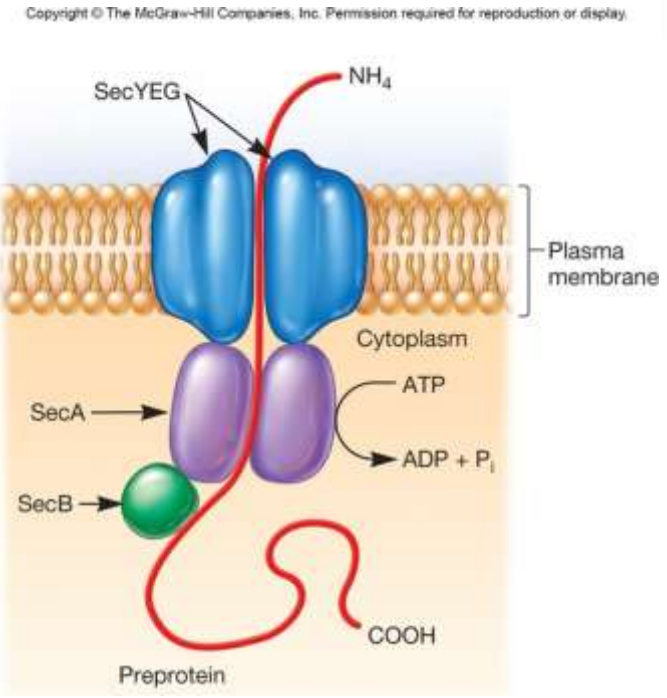
Sec-Dependent Pathway

- Also called general secretion pathway
  - highly conserved in all domains
- Translocates proteins from cytoplasm across or into plasma membrane
- Secreted proteins synthesized as preproteins having amino-terminal signal peptide
  - **signal peptide** delays protein folding



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Sec-Dependent Pathway

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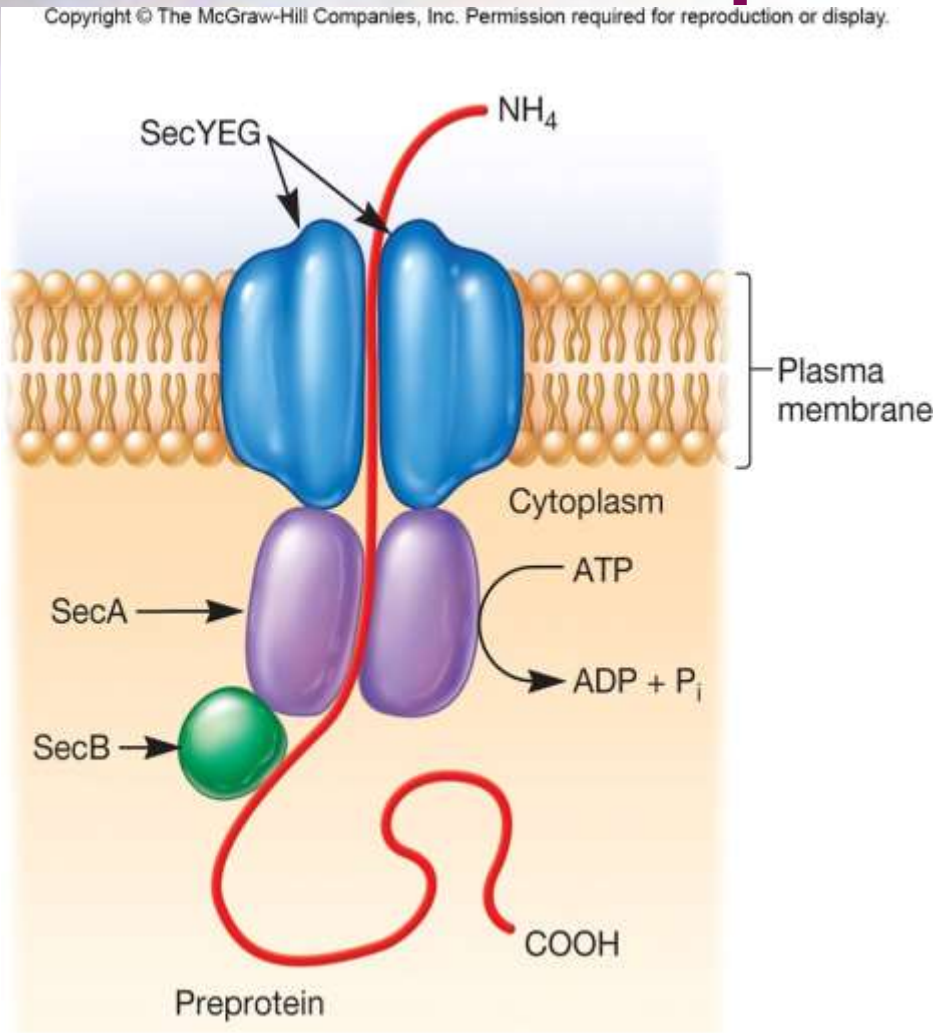

- **secY, secE, and secG form a channel in the membrane**
- **secA translocates preprotein through the plasma membrane**
- When preprotein emerges from(يخرج من) plasma membrane a signal peptidase removes the

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Other Protein Secretion Pathways

- **Type I secretion systems**
  - related to ABC transport systems
  - Gram-positive/Gram-negative bacteria, and *Archaea*
  - Secretion of toxins, proteases, other proteins

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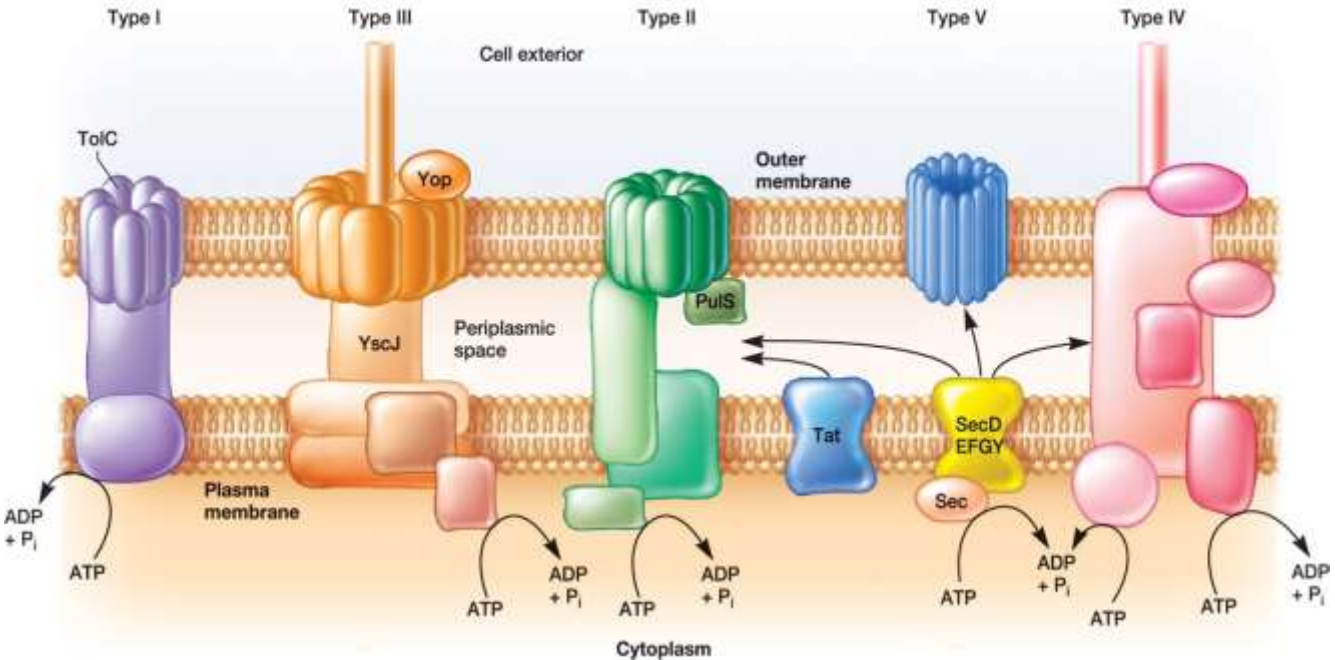

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Tat System

- Protein translocation **system in *Bacteria* and some archaea**
- Moves across plasma membrane
- Tat pathway translocated **folded proteins with "twin" arginine residues in their signal sequence**
- Works with Type II secretion system

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Other Protein Secretion Pathways

- **Type IV secretion system**
  - –secrete proteins
  - –secrete DNA from donor to recipient bacterium during conjugation
  - –found in both Gram-positive and Gram- negative

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Protein Secretion in Gram-Negative Bacteria

- Six proteins secretion systems identified
- **Types I and IV also in Gram-positives**
- **Types II, III, and V are unique to Gram-negatives**
  - **most secrete virulence factors**

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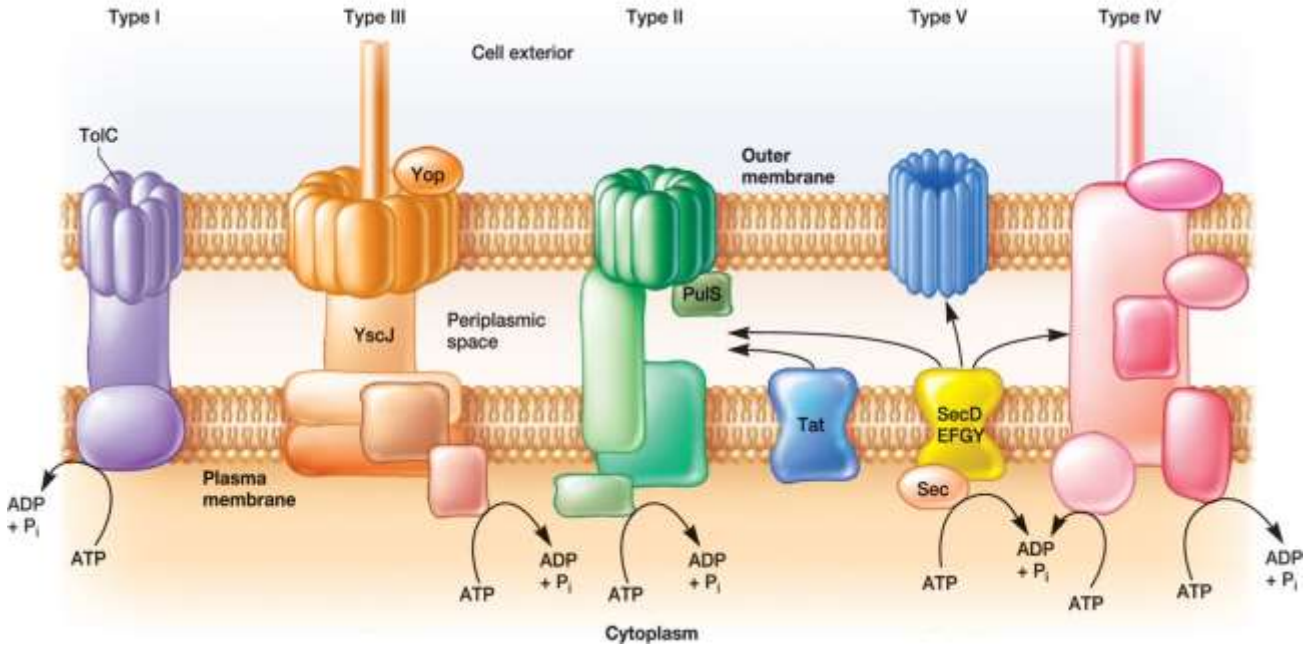

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Gram-Negative Bacteria Secretion Systems

- **Types I and III are sec independent**
  - **forms injectisomes**
  - **transports virulence factors and other proteins**
- **Type V are sec-dependent**
  - **autotransporters – transport themselves out**
- Type VI are similar to bacteriophage genome injection systems

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