DNA TRANSCRIPTION AND TRANSLATION

ASSIST PROF. DR. MUJAHID KH. ALI

General Objectives:

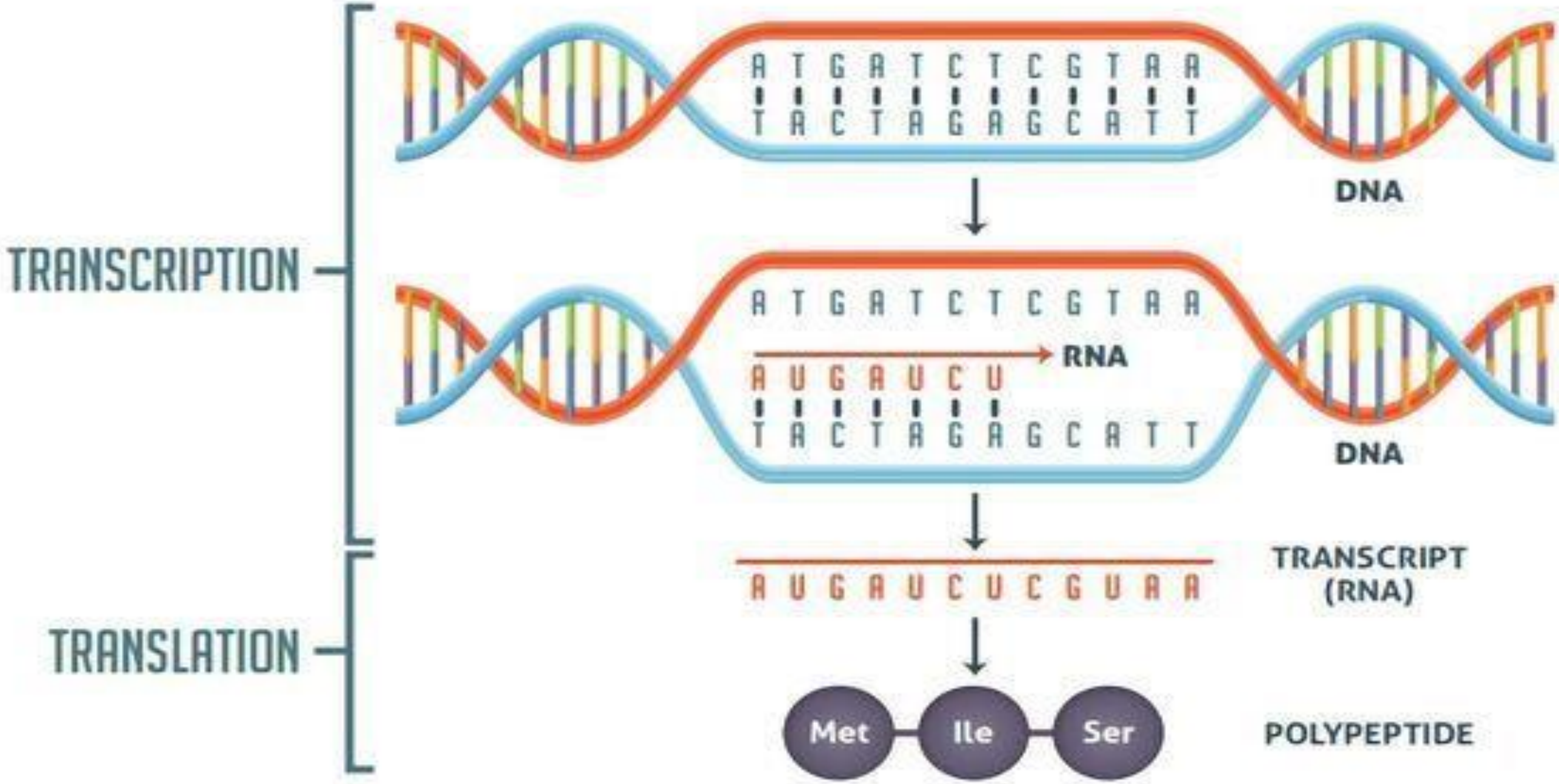
1. To understand the two-step process of converting a gene into a protein (Transcription and Translation).
2. To explain the process of transcription: principles, steps (Initiation, Elongation, Termination), and the role of RNA Polymerase.
3. To differentiate between the template strand and the coding strand.
4. To explain the process of translation: principles, steps (Initiation, Elongation, Termination), and the role of the ribosome, tRNA, and mRNA.
5. To understand the concept of codons and anticodons and how they determine the amino acid sequence.

Basically, a gene is used to build a protein in a two-step process:

Step1-Transcription is the first step of gene expression. During this process, the DNA sequence of a gene is copied into RNA.

Before transcription can take place, the DNA double helix must unwind near the gene that is getting transcribed.

The region of opened-up DNA is called a **transcription bubble**.


TRANSCRIPTION
TRANSLATION
A T G A T C T C G T A A
T A C T A G A G C A T T
DNA
A T G A T C T C G T A A
RNA
A U G A U C U
T A C T A G A G C A T T
DNA
A U G A U C U C G U A A
TRANSCRIPT (RNA)
Met
Ile
Ser
POLYPEPTIDE

The complementary RNA is created from 5’-3’ direction. Although The DNA uses one of the two strands as a template; this strand is called the **template strand**. The RNA product is complementary to the template strand and is almost identical to the other DNA strand, called the **non-template** (or **coding**) **strand**. However, there is one important difference: in the newly made RNA, all of the T nucleotides are replaced with U nucleotides.

The site on the DNA from which the first RNA nucleotide is transcribed is called the +1 plus1 site, or the **initiation site**.

Nucleotides that come before the initiation site are given negative numbers and said to be **upstream**. Nucleotides that come after the initiation site are marked with positive numbers and said to be **downstream**.

If the gene that's transcribed encodes a protein (which many genes do), the RNA molecule will be read to make a protein in a process called translation.

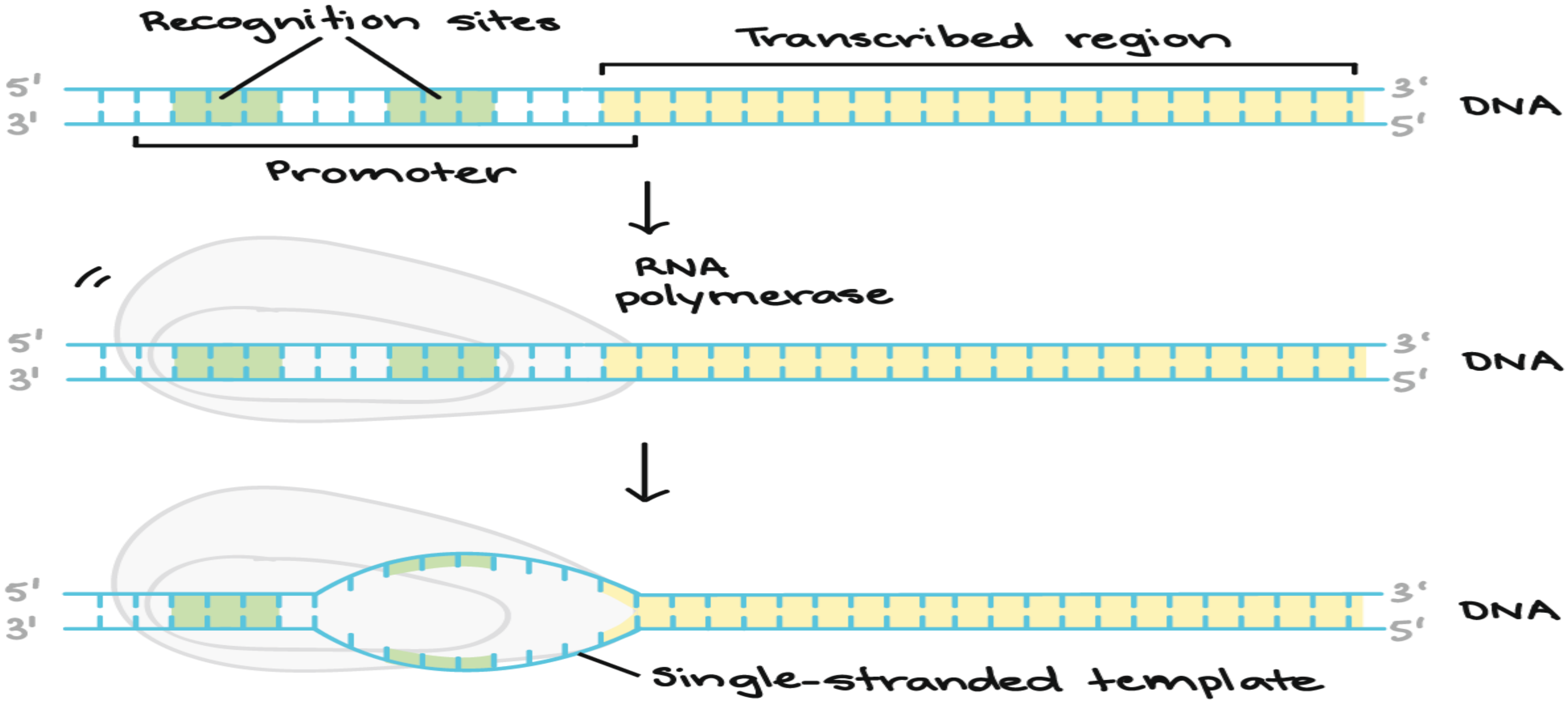
The process of Transcription;

There are three main steps to the process of DNA transcription;

1- RNA polymerase binds to DNA

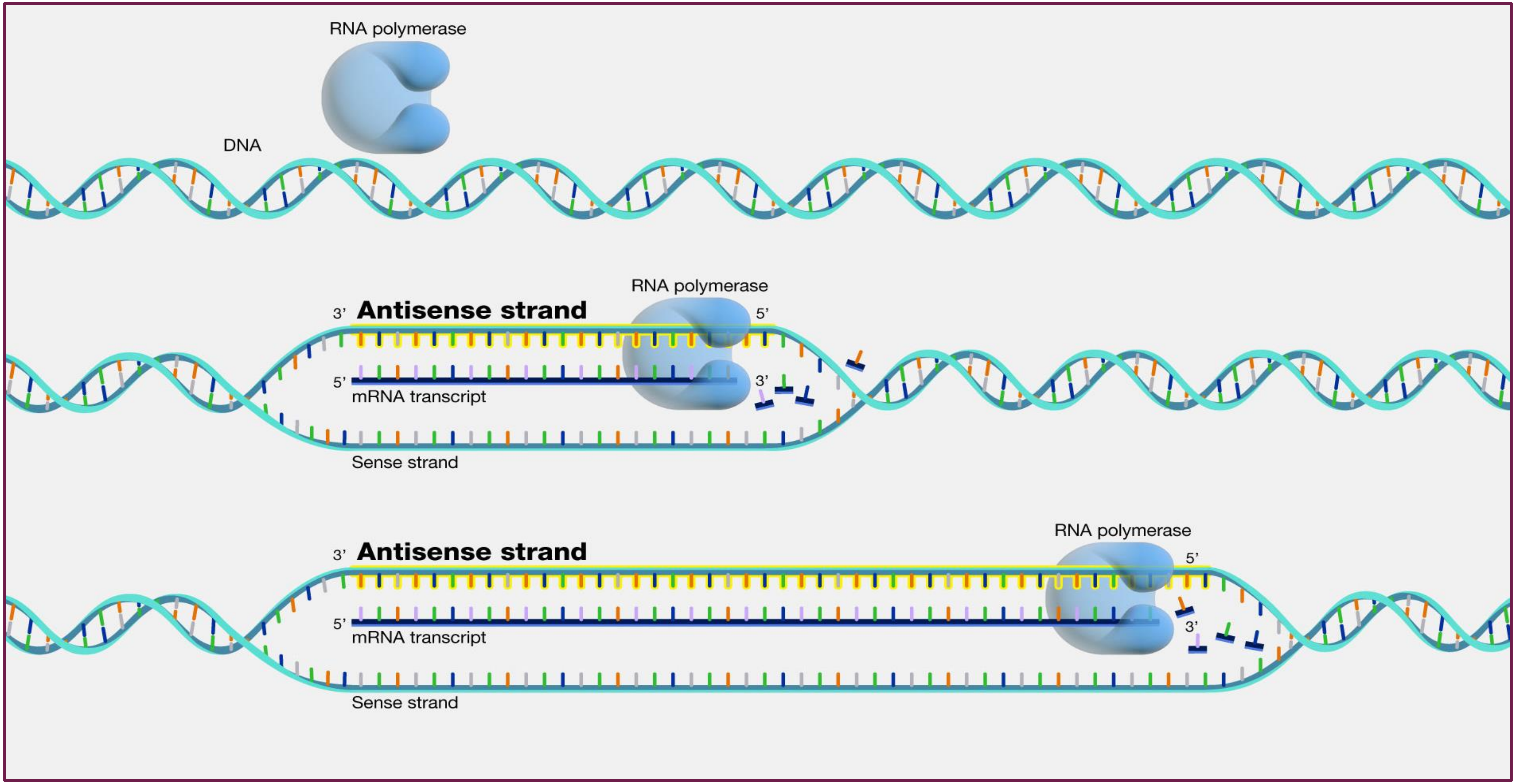
(Initiation); An enzyme called RNA polymerase transcribes the DNA.

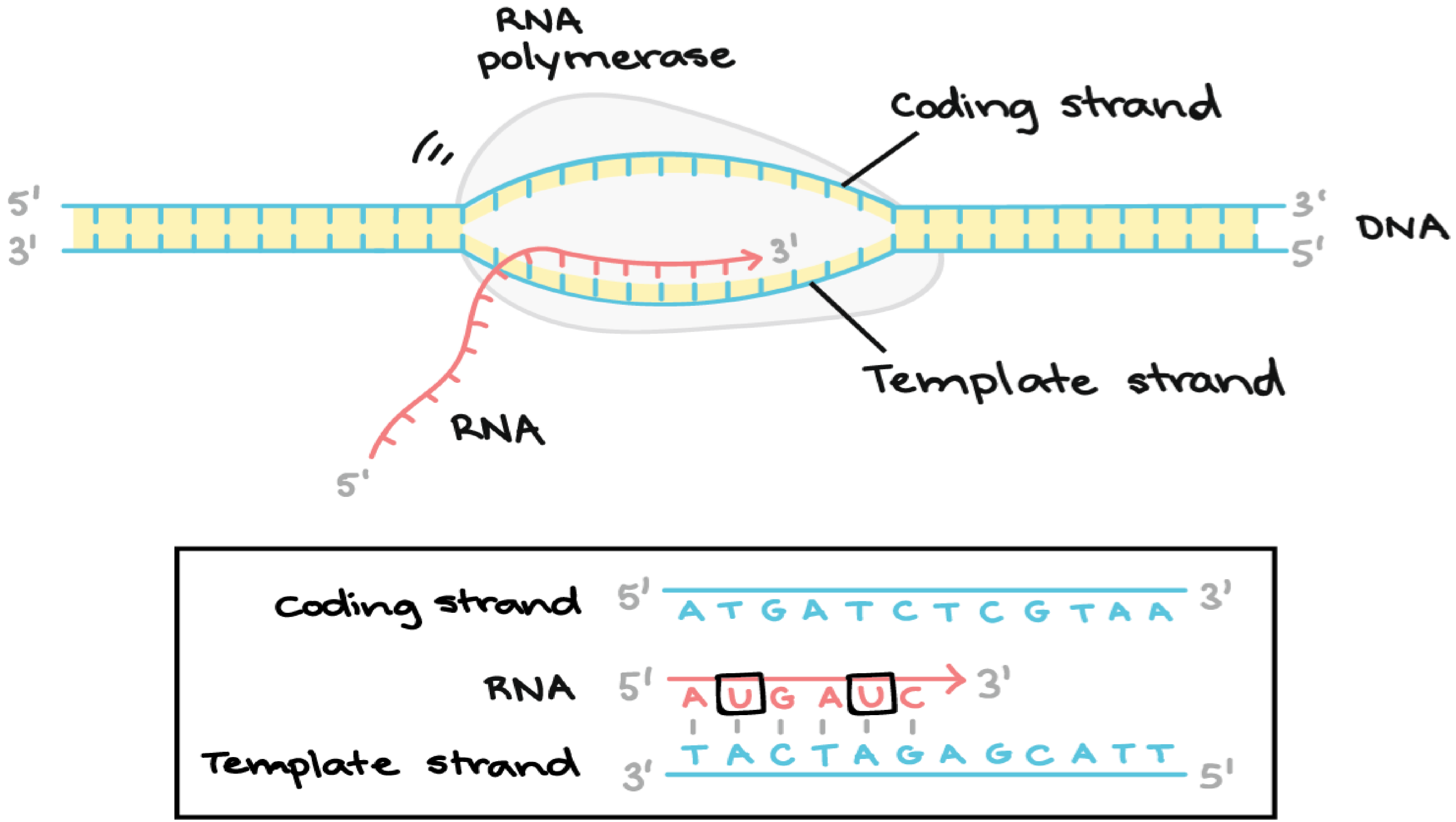
Specific nucleotide sequences tell RNA polymerase where to begin and where to end. RNA polymerase attaches to the DNA at a specific area called the **promoter region**


Recognition sites
Transcribed region
5'
3'
3'
5'
DNA
Promoter
RNA
polymerase
5'
3'
3'
5'
DNA
5'
3'
3'
5'
DNA
Single-stranded template

(Elongation) certain protein called transcription factor (with the aid of **helicase enzyme**) unwind the DNA strand- by breaking the hydrogen bonds between complementary DNA nucleotide- and allow RNA polymerase to transcript only a single strand of DNA into a single stranded RNA polymer called messenger RNA (mRNA). RNA polymerase adds matching RNA nucleotides that are paired with complementary DNA nucleotides of one DNA strand. The strand that serves as the template is called the **Antisense strand** . The strand that is not transcribed is called the **sense stand**

(Termination); RNA polymerase moves along the DNA until it reaches a terminator sequence. At that point RNA polymerase releases the mRNA polymer by breaking the hydrogen bonds of the untwisted RNA+DNA helix, freeing the newly synthesized RNA strand, then the RNA polymerase detaches from the DNA.


RNA polymerase
DNA
RNA polymerase
3'
Antisense strand
5'
5'
3'
mRNA transcript
Sense strand
RNA polymerase
3'
Antisense strand
5'
5'
3'
mRNA transcript
Sense strand


RNA polymerase
Coding strand
5'
3'
3'
5'
DNA
3'
Template strand
RNA
5'
Coding strand
5' A T G A T C T C G T A A 3'
RNA
5' A U G A U C 3'
Template strand
3' T A C T A G A G C A T T 5'

Translation:

Translation: The big picture

Translation involves "decoding" a messenger RNA (mRNA) and using its information to build a **polypeptide**, or chain of amino acids. For most purposes, a polypeptide is basically just a protein (with the technical difference being that some large proteins are made up of several polypeptide chains).

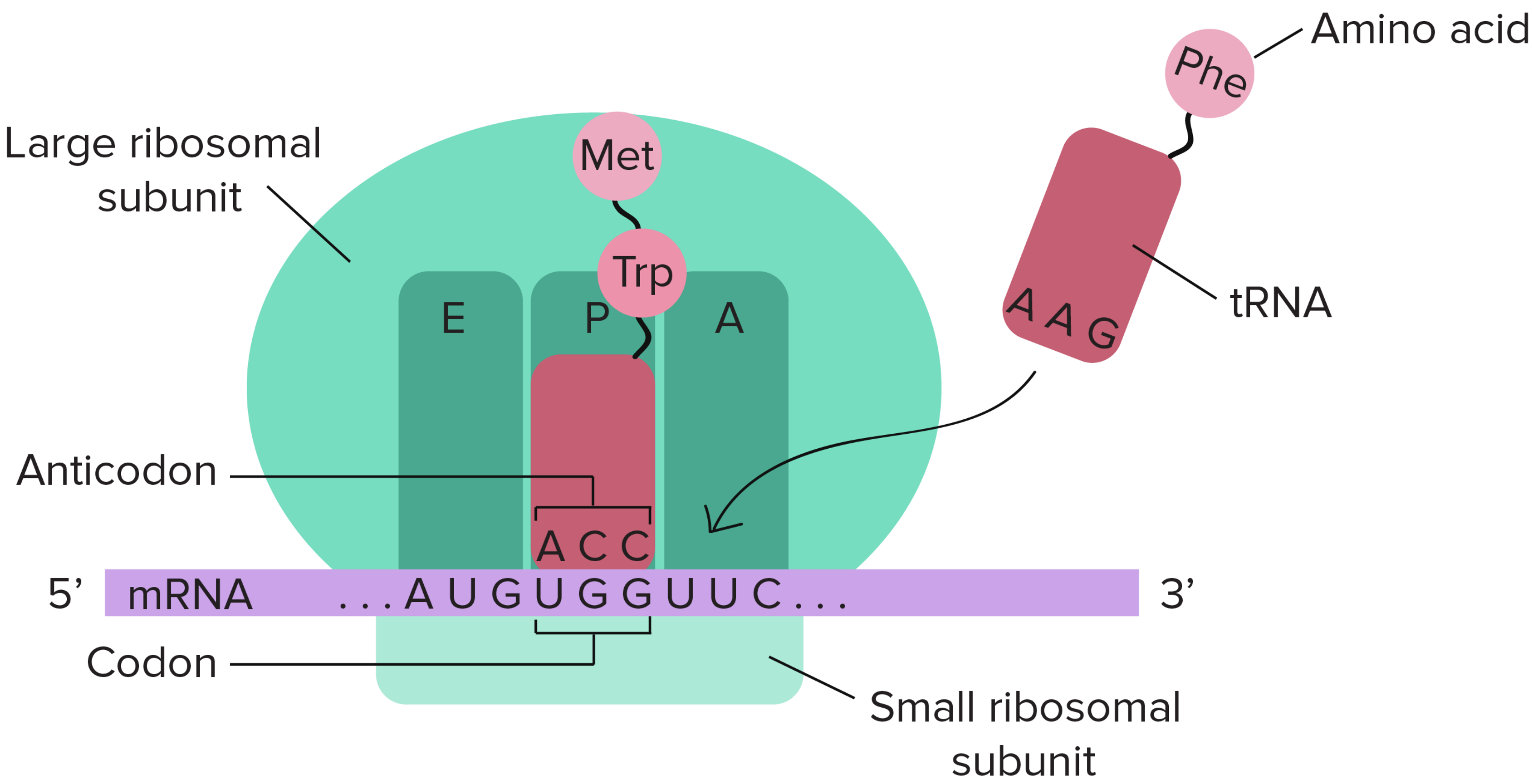
Codons to amino acids

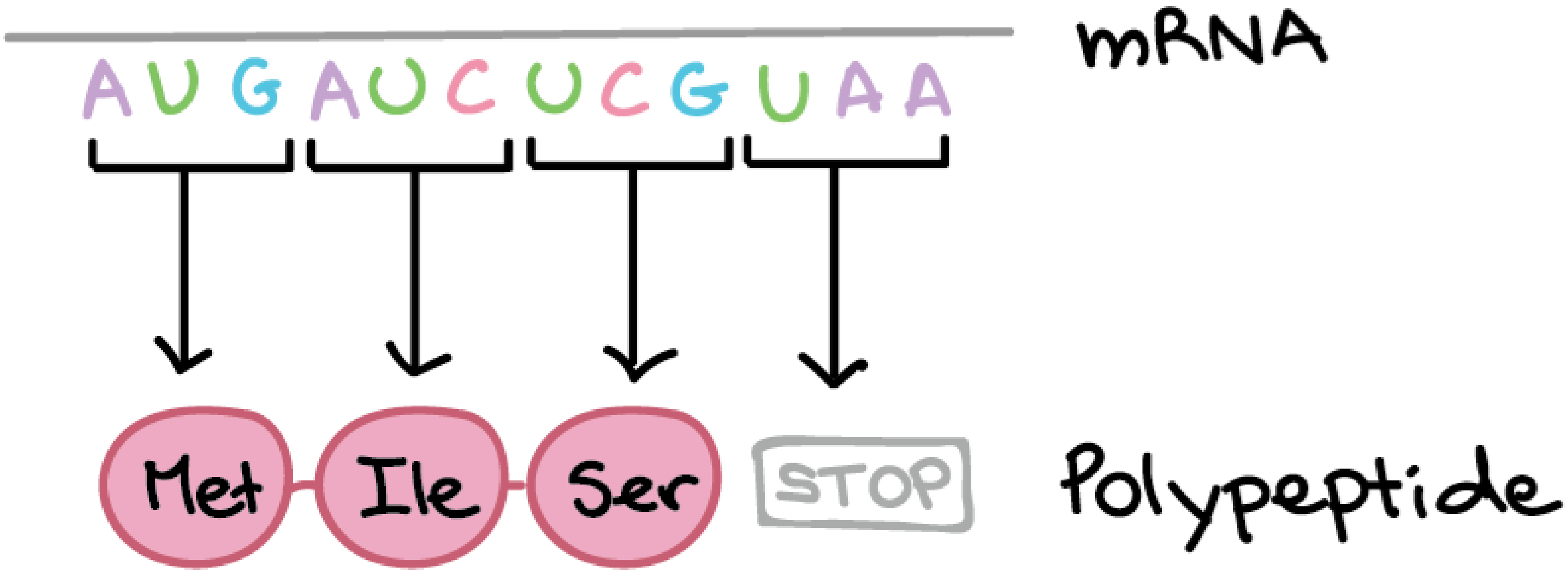
In translation, the codons of an mRNA are read in order (from the 5' end to the 3' end) by molecules called **transfer RNAs**, or **tRNAs**.

Each tRNA has an **anticodon**, a set of three nucleotides that binds to a matching mRNA codon through base pairing. The other end of the tRNA carries the amino acid that's specified by the codon.

Step 2: DNA

In this stage, the mRNA is "decoded" to build a protein that contains a specific series of amino acids.


Amino acid
Phe
Large ribosomal subunit
Met
Trp
E
P
A
tRNA
A A G
Anticodon
A C C
5'
mRNA
. . . A U G U G G U U C . . .
3'
Codon
Small ribosomal subunit



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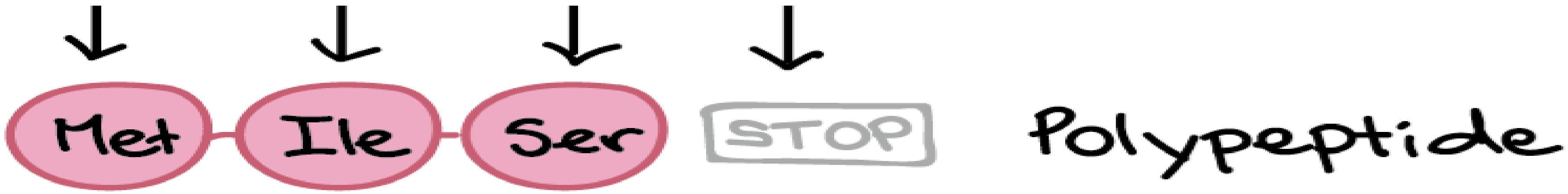


•**Initiation** ("beginning"): in this stage, the ribosome gets together with the mRNA and the first tRNA so translation can begin.

•**Elongation** ("middle"): in this stage, amino acids are brought to the ribosome by tRNAs and linked together to form a chain.

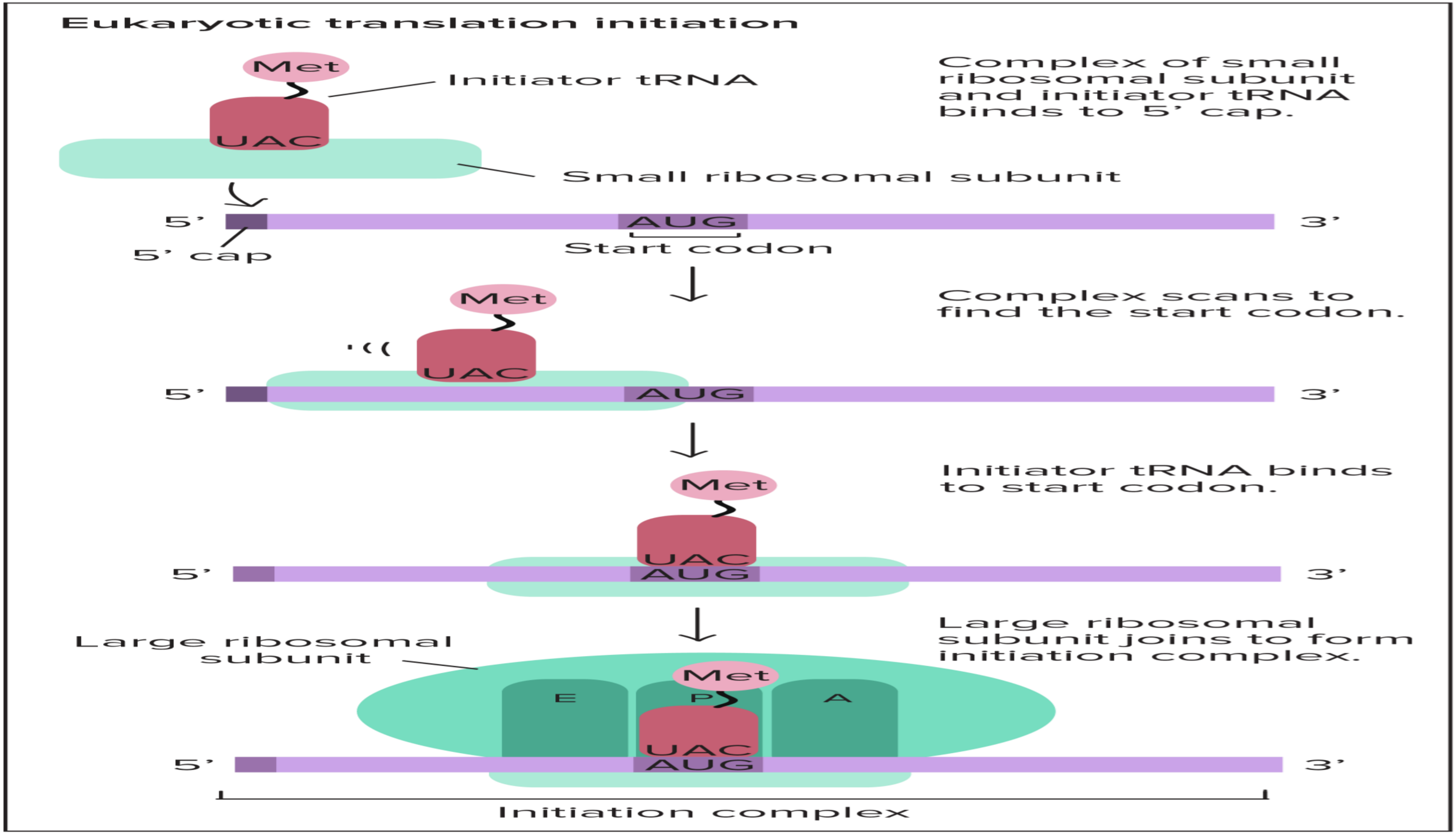
•**Termination**("end"): in the last stage, the finished polypeptide is released to go and do its job in the cell.

Let’s take a closer look at how each stage works.

1.Initiation

- A ribosome (which comes in two pieces, large and small)
- An mRNA with instructions for the protein we'll build
- An "initiator" tRNA carrying the first amino acid in the protein, which is almost always methionine (Met) .

Inside our cells (eukaryotes), translation initiation goes like this: first, the tRNA carrying methionine attaches to the small ribosomal subunit. Together, they bind to the 5' end of the mRNA by recognizing the 5' GTP (Guanosine-5'-triphosphate) cap (added during processing in the nucleus). Then, they "walk" along the mRNA in the 3' direction, stopping when they reach the start codon (often, but not always, the first AUG).



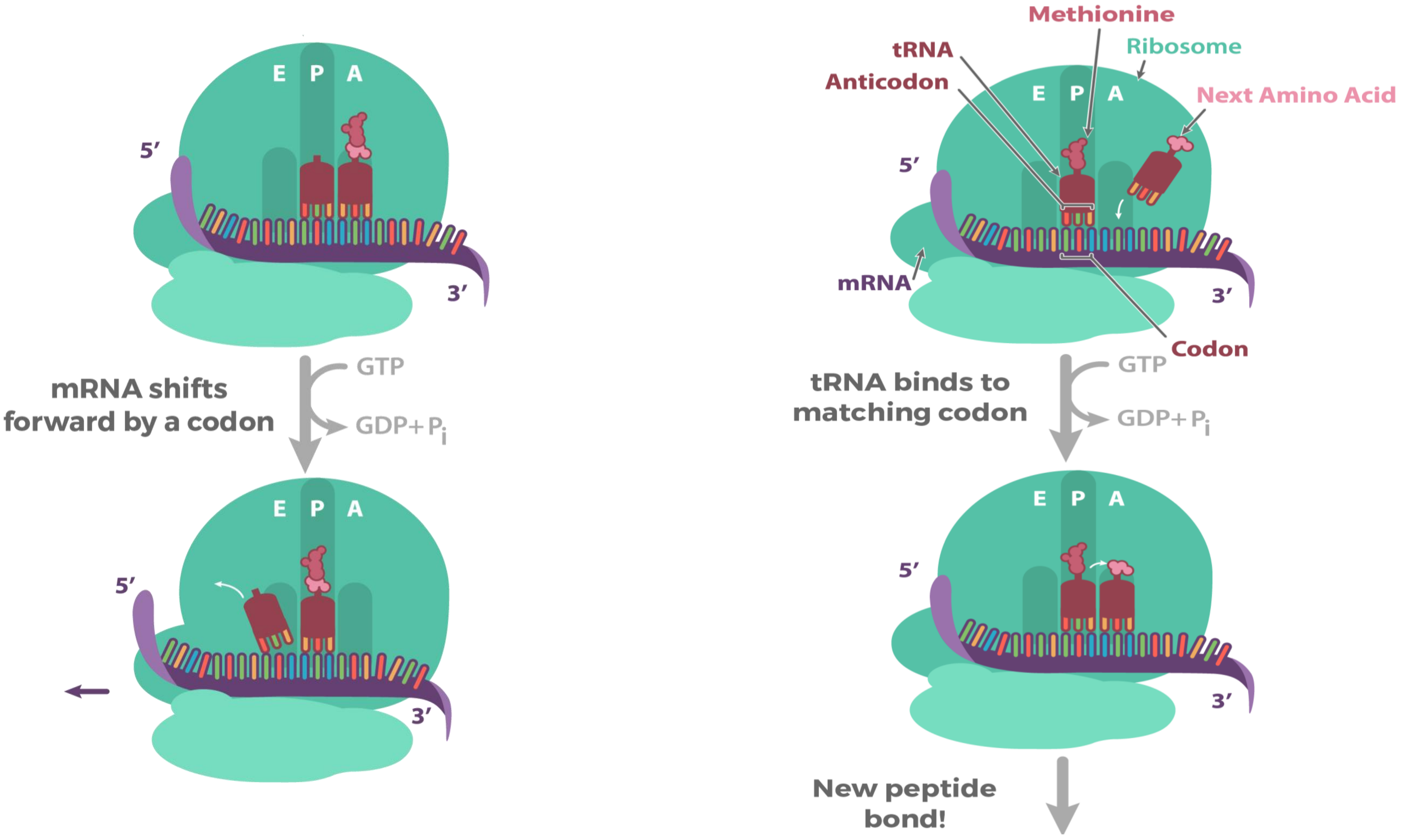
Eukaryotic translation initiation
Met
Initiator tRNA
UAC
Complex of small ribosomal subunit and initiator tRNA binds to 5' cap.
Small ribosomal subunit
5'
AUG
3'
5' cap
Start codon
Met
Complex scans to find the start codon.
UAC
5'
AUG
3'
Met
Initiator tRNA binds to start codon.
UAC
AUG
5'
3'
Large ribosomal subunit
Large ribosomal subunit joins to form initiation complex.
Met
E
P
A
UAC
AUG
5'
3'
Initiation complex

2. Elongation

Elongation is when the polypeptide chain gets longer. But how does the chain actually grow? To find out, let's take a look at the first round of elongation—after the initiation complex has formed, but before any amino acids have been linked to make a chain.

Our first, methionine-carrying tRNA starts out in the middle slot of the ribosome, called the P site. Next to it, a fresh codon is exposed in another slot, called the A site. The A site will be the "landing site" for the next tRNA, one whose anticodon is a perfect (complementary) match for the exposed codon.

First round of elongation

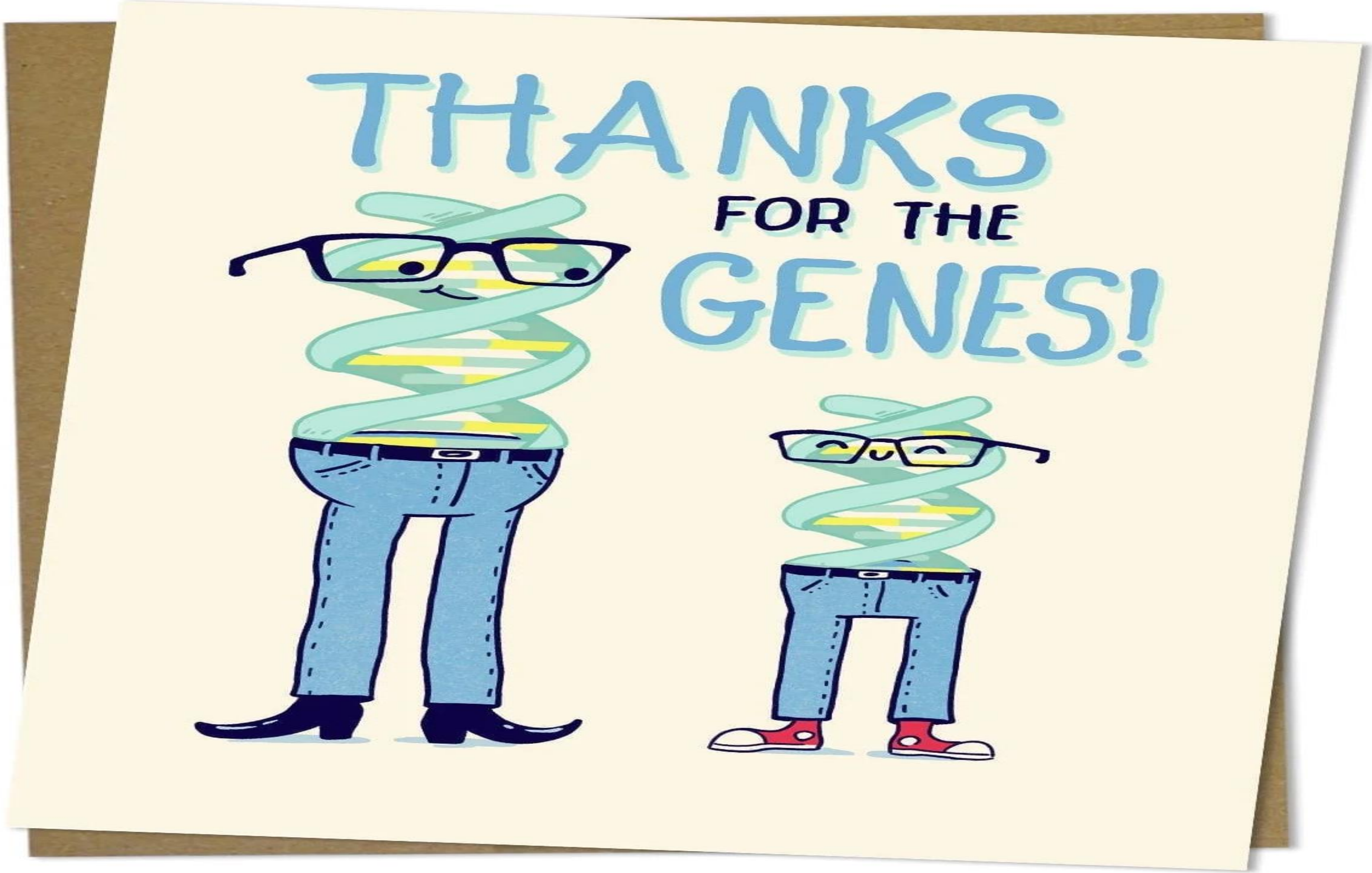


Once the matching tRNA has landed in the A-site, it's time for the action: that is, the formation of the **peptide bond** that connects one amino acid to another. This step transfers the methionine from the first tRNA onto the amino acid of the second tRNA in the A site.

But...odds are we may want a longer polypeptide than two amino acids. How does the chain continue to grow? Once the peptide bond is formed, the mRNA is pulled onward through the ribosome by exactly one codon. This shift allows the first; empty tRNA to drift out via the E ("exit") site. It also exposes a new codon in the A site, so the whole cycle can repeat.

3. Termination: is the stage in which the finished polypeptide chain is released. It begins when a stop codon (UAG, UAA, or UGA) enters the ribosome, triggering a series of events that separate the chain from its tRNA and allow it to drift out of the ribosome.

After termination, the polypeptide may still need to fold into the right 3D shape, undergo processing (such as the removal of amino acids), get shipped to the right place in the cell, or combine with other polypeptides before it can do its job as a functional protein.


THANKS
FOR THE
GENES!