4.4: Gene Regulation

Chapter 16

Learning objectives

- **Understand why cells regulate genes** - to save energy, specialize cells, and respond to changes

- **Know the 4 levels of gene regulation:**
- Transcriptional (making mRNA)
- Post-transcriptional (processing mRNA)
- Translational (making protein)
- Post-translational (modifying protein)

- **Explain how transcription is controlled:**
- Chromatin: open (ON) vs. closed (OFF)
- Transcription factors: activators (turn ON) and repressors (turn OFF)
- Epigenetics: changes that don't alter DNA sequence

- **Explain how identical DNA → different cells** through gene regulation

- **Understand alternative splicing** - one gene makes multiple proteins

Two cats with <u>identical DNA sequences</u> for the pigment gene

- One is solid black
- One is black and white spotted

Question: The DNA is the same... so what's different?

Answer: Not all DNA is "read" the same way in every cell

Previously: How cells decide WHICH genes to read and HOW they read them

Today: How do cells with identical genes become so different? Gene REGULATION!

ICA Q1: You have liver cells and skin cells. Both have genes for:

- **Insulin** (made in pancreas)
- **Hemoglobin** (made in red blood cells)
- **Keratin** (made in skin)

Which genes should be "on" in each cell type? Why?

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- **Insulin** (made in pancreas)
- **Hemoglobin** (made in red blood cells)
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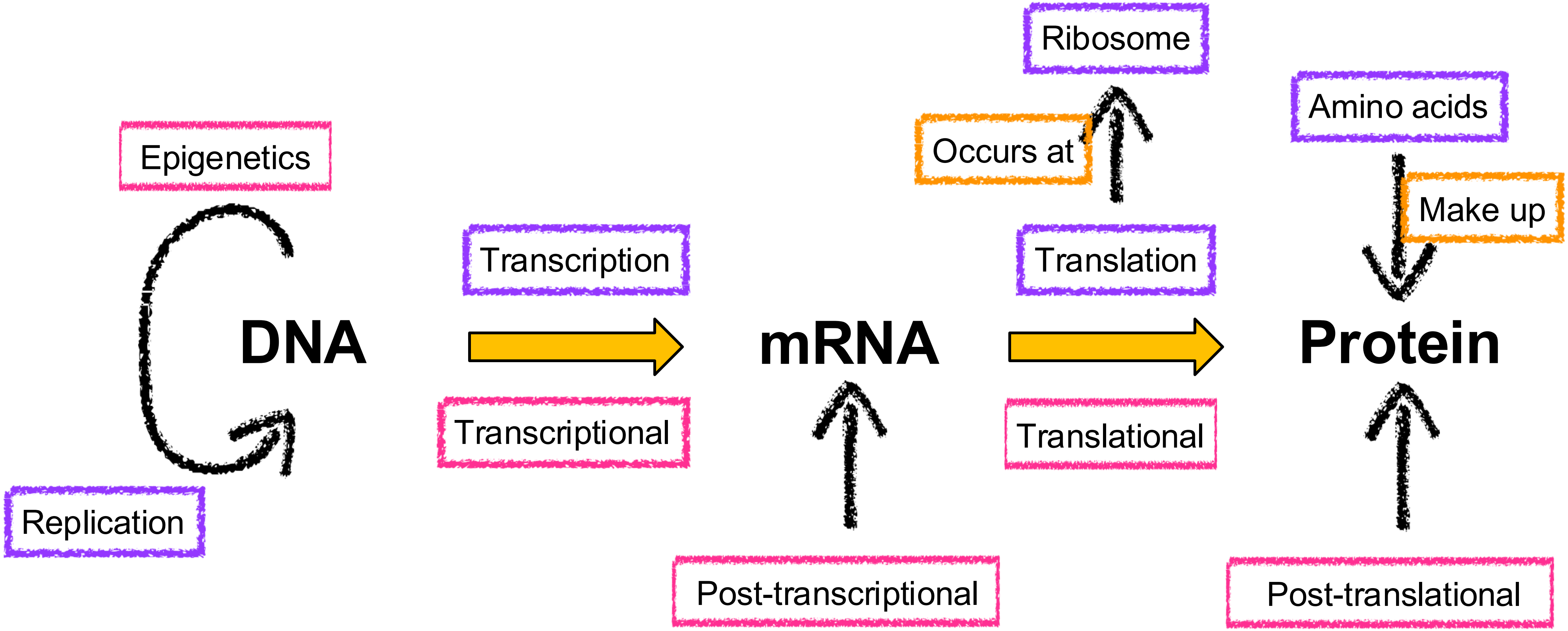
Which genes should be "on" in each cell type? Why?

Liver – none of these
Skin - keratin

Gene Expression Can Be Regulated at Multiple Steps

- Transcriptional control (making mRNA & Epigenetics)
- Post-transcriptional control (processing mRNA)
- Translational control (making protein)
- Post-translational control (modifying protein)



ICA Q2: Why do cells need gene regulation? Select all that apply:

A) To save energy by not making unneeded proteins
B) To allow cell specialization
C) To respond to environmental changes
D) To replicate DNA faster

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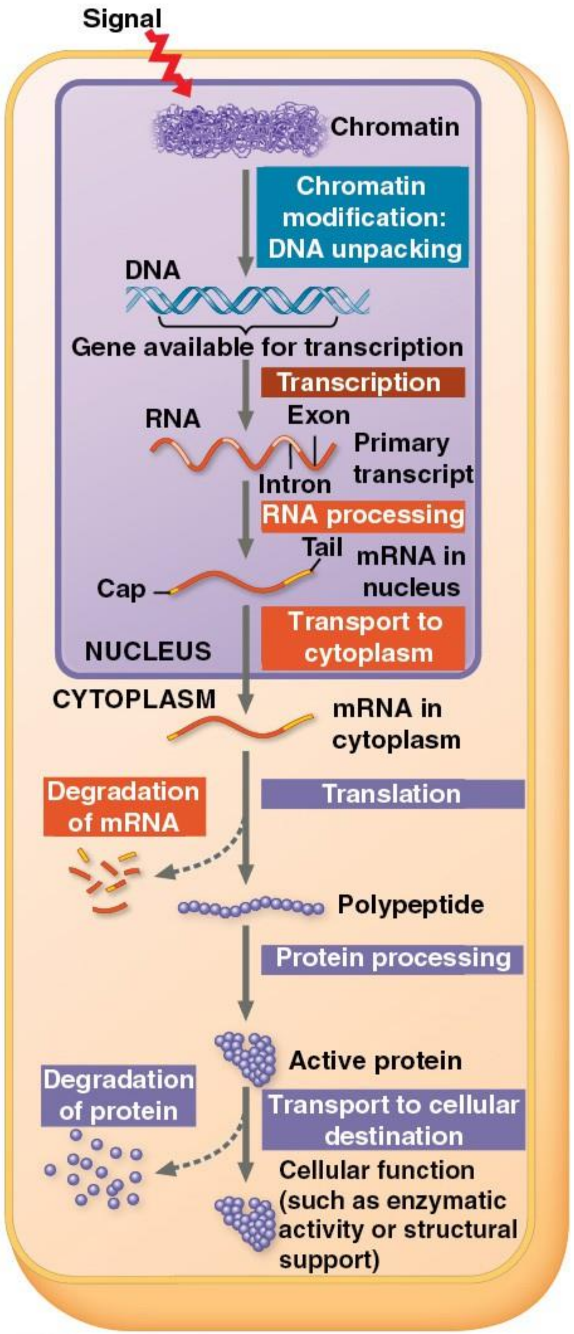


- Almost all the cells in an organism contain an identical genome
- All organisms must regulate which genes are expressed at any given time.
- Genes are turned on and off in response to signals from their external and internal environments
- In multicellular organisms, regulation of gene expression is essential for cell specialization
- Regulation maintains efficiency
  - **Energy** – expressing all genes would require a massive amount of energy
  - **Space** – cells are kept to a manageable size
  - **Time** – genes can be expressed as needed and more rapidly.

Transcriptional regulation controls IF and HOW MUCH mRNA is made

Three main levels:

1. <u>Chromatin structure:</u> is the chromatin open or closed?

Heterochromatin	Euchromatin
Tightly packed → Genes OFF	Loosely packed → Genes accessible/ON

2. <u>Transcription factors:</u> proteins that influence whether or not transcription starts

Repressors	Activators
Decrease transcription	Increase transcription

3. <u>Epigenetics:</u> any process that alters gene activity without changing the DNA sequence

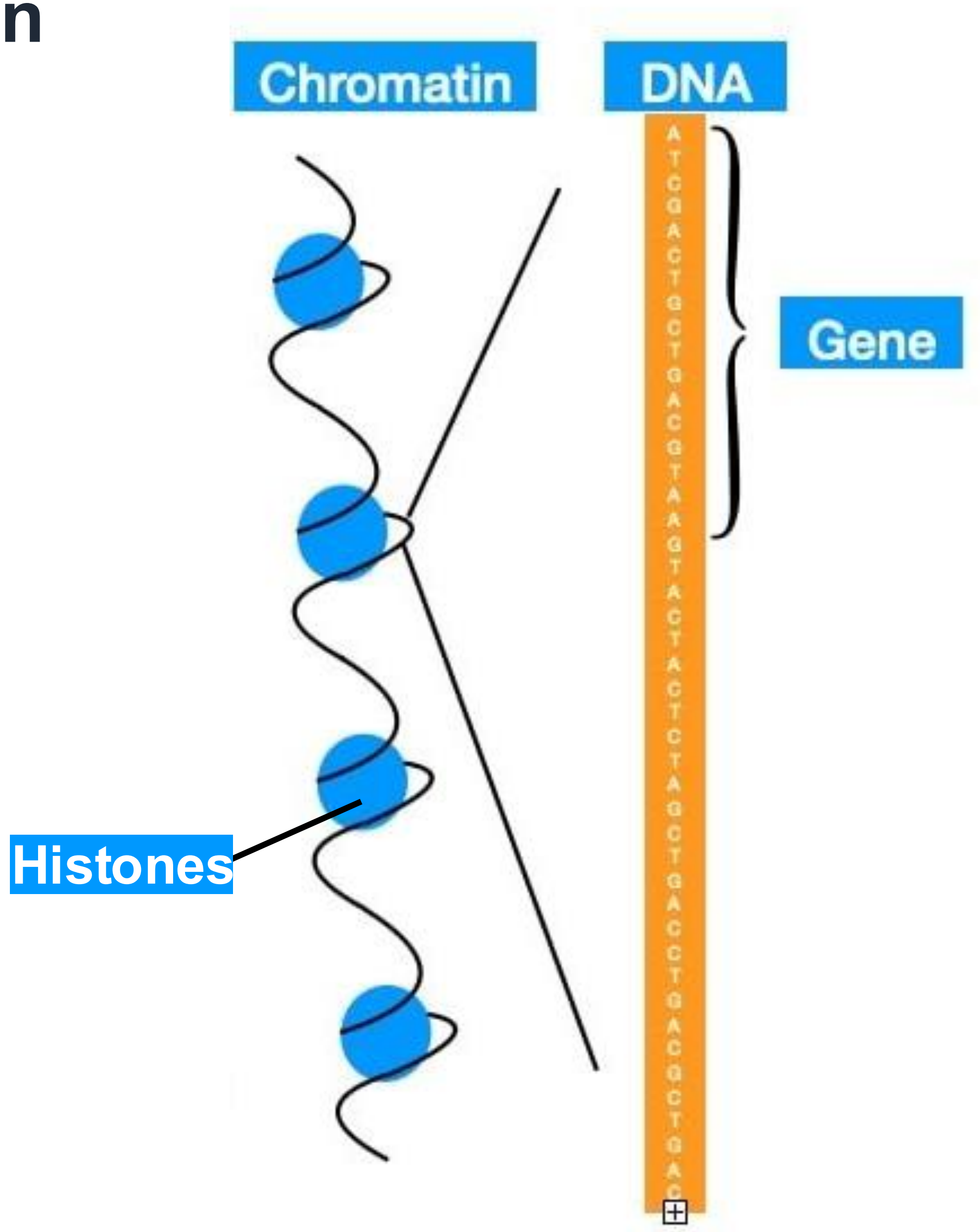
1. <u>Chromatin structure:</u> is the chromatin open or closed?

DNA Packaging Creates a Regulation Problem

- DNA must fit inside nucleus (2 meters into ~10 μm!)
- DNA wraps around histone proteins
- DNA IS wrapped around histone proteins like beads on a string

Problem: wrapped DNA is not accessible

Solution: cells can control how tightly DNA is packed

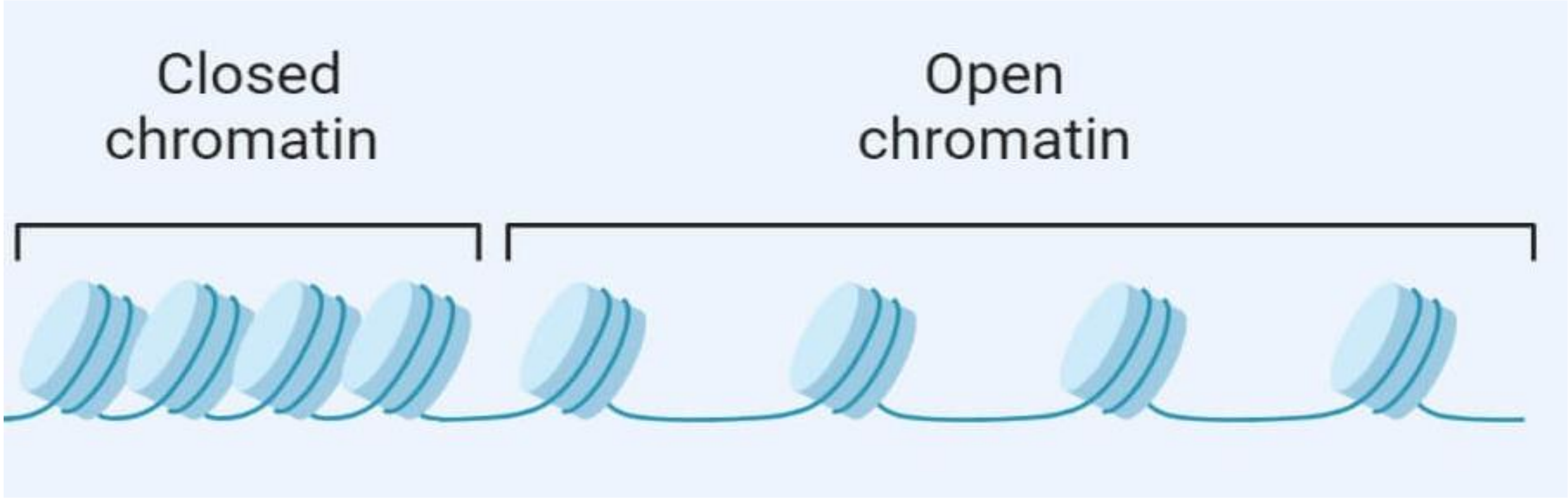


1. Chromatin structure: is the chromatin open or closed?

Two States of Chromatin

Heterochromatin: tightly packed, genes **OFF**
- DNA wrapped tightly around histones
- Proteins cannot access DNA
- RNA polymerase cannot bind

Euchromatin: loosely packed, genes accessible, **ON**
- DNA more open and available
- Proteins can bind
- Genes can be transcribed



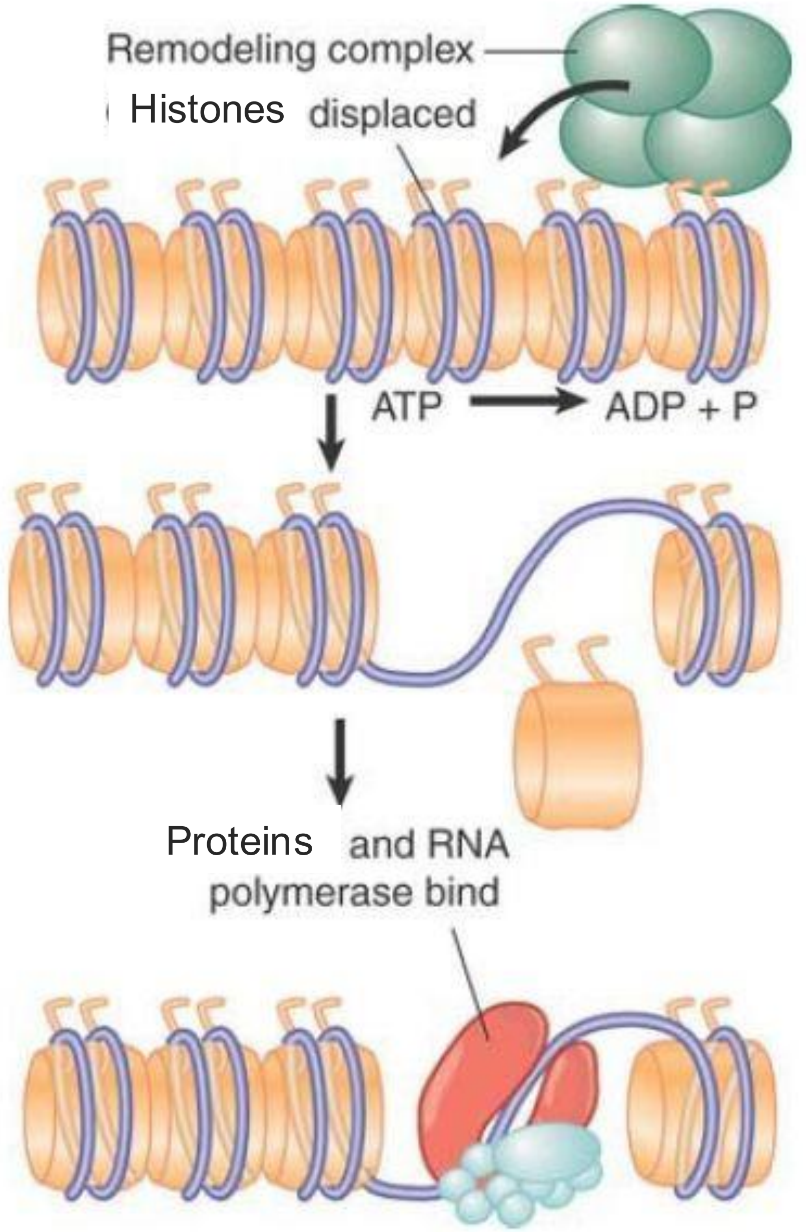
1. <u>Chromatin structure:</u> is the chromatin open or closed?

Chromatin Remodeling is transcriptional regulation

- Special protein complexes can physically move histones
- Requires energy (ATP)
- Can slide histones along DNA
- Can temporarily remove histones
- "Opens up" DNA so regulatory proteins can access it
- First step in gene activation

ICA Q3: A gene is located in tightly packed heterochromatin. Even though all the correct proteins for transcription are present in the nucleus, the gene is not expressed. Why?

A) The transcription proteins are defective
B) RNA polymerase is missing
C) Transcription factors cannot physically reach the DNA
D) The DNA sequence has been deleted



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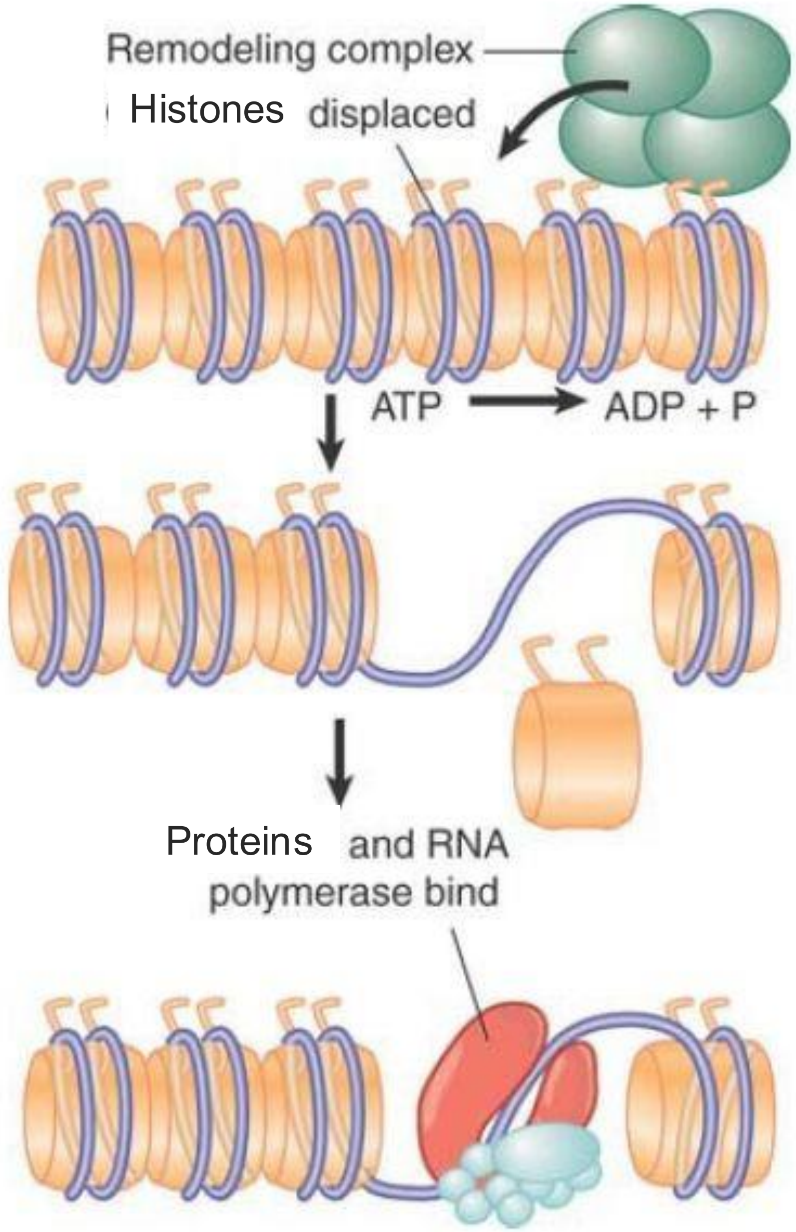


2. Transcription factors: proteins that influence whether or not transcription starts

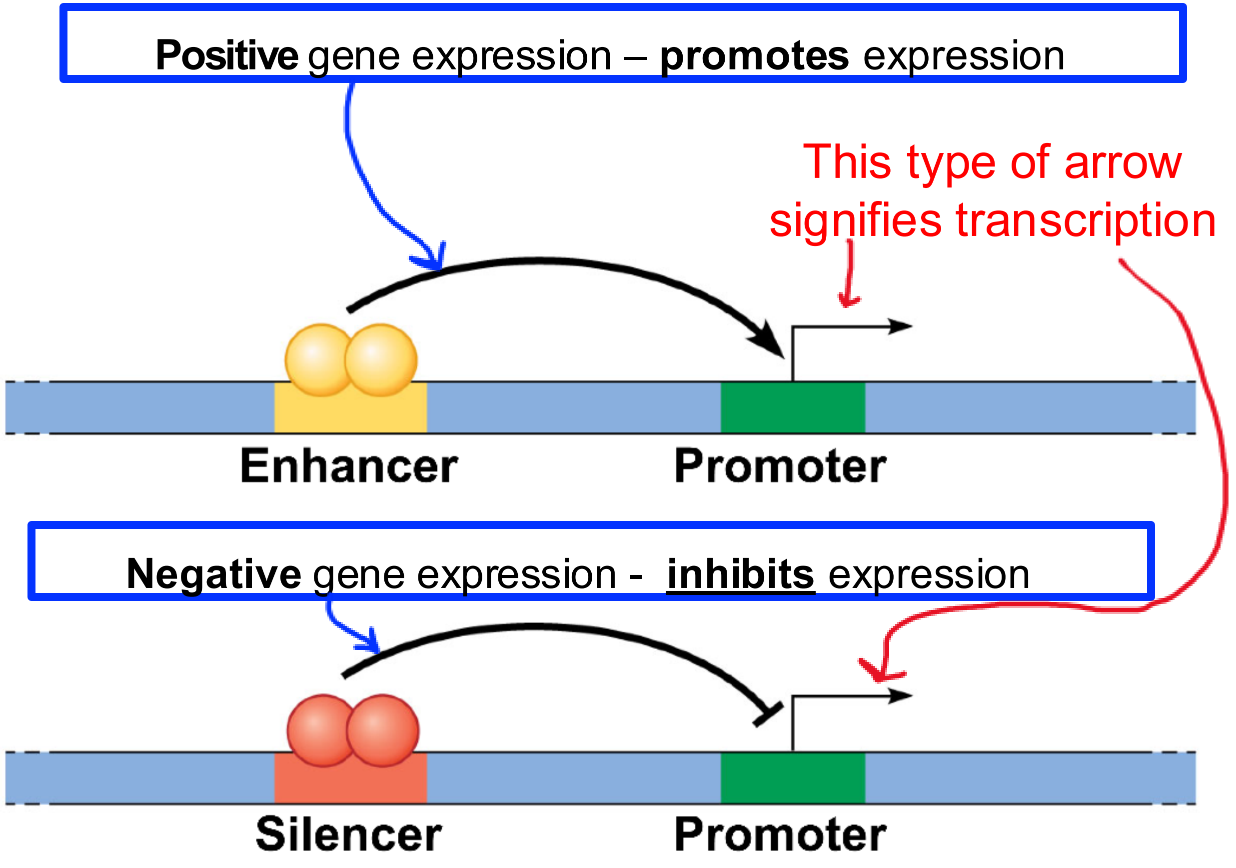
The Promoter and Regulatory Sequences

- **Promoter**: DNA sequence where RNA polymerase binds (near start of gene)
- **Enhancers**: DNA sequences that increase transcription
- **Silencers**: DNA sequences that decrease transcription

Key point: these are DNA sequences, not proteins. They are binding sites for regulatory proteins

Transcription Factors - The Key Regulatory Proteins

- Proteins that bind to regulatory DNA sequences
- Have two functional parts:
    - **DNA-binding domain**: recognizes and binds specific DNA sequence
    - **Regulatory domain**: affects transcription rate
- Two main types:
    - **Activators**
    - **Repressors**



ICA Q4: Predict which type of transcription factor binds to the enhancer. To the activator?

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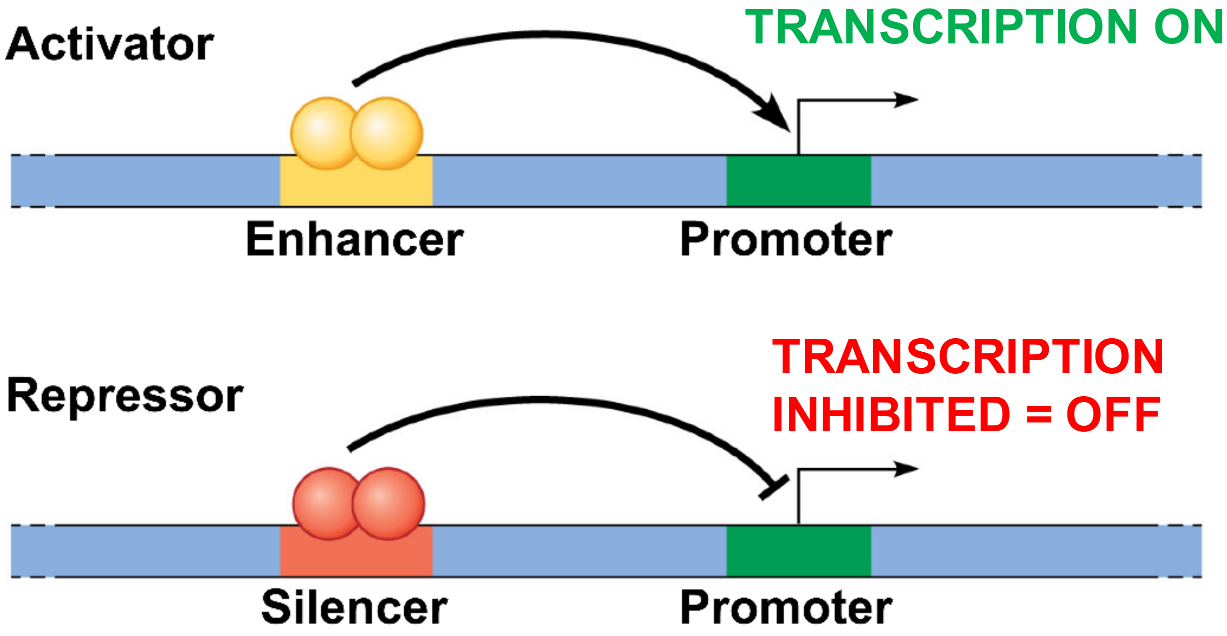

ICA Q4: Predict which type of transcription factor binds to the enhancer. To the activator?

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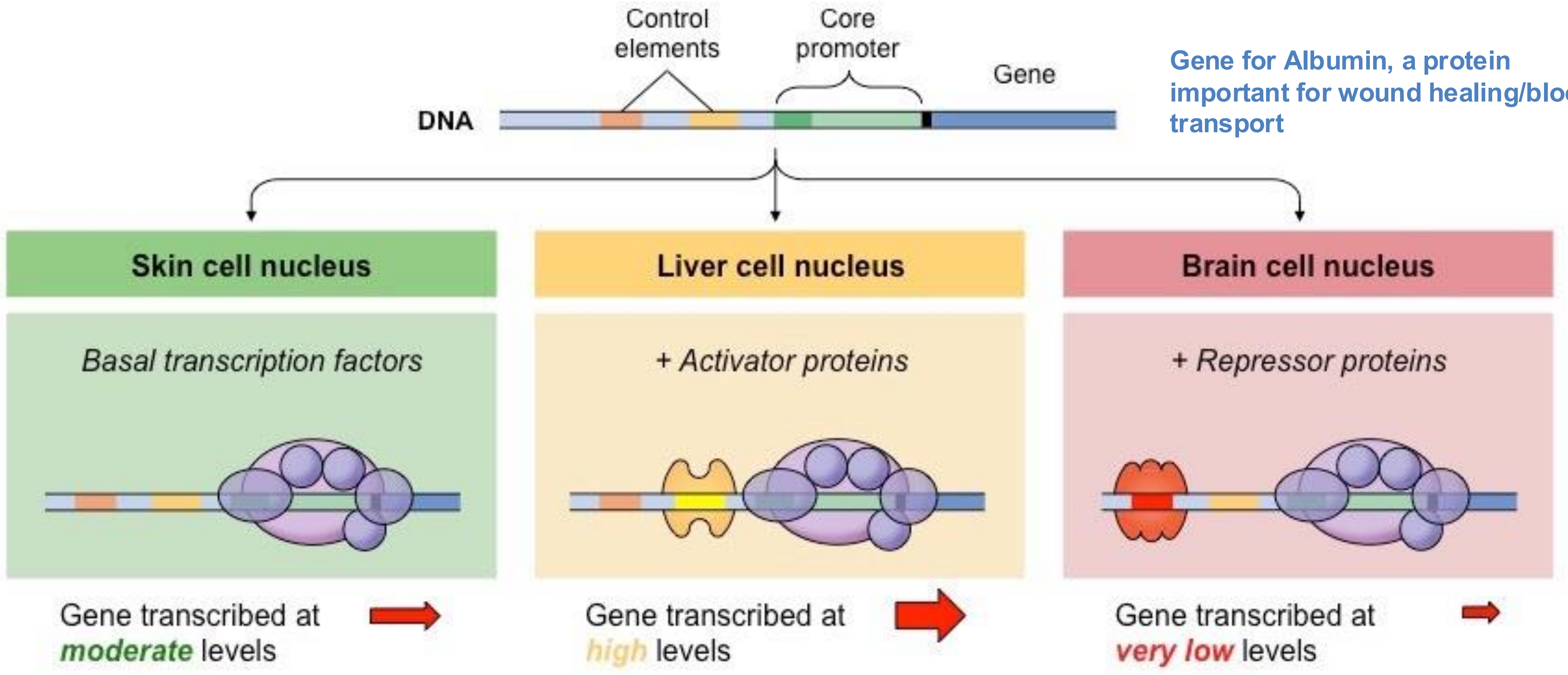
BASAL transcription factors = general transcription factors, which are the essential proteins needed to initiate transcription by recruiting RNA polymerase



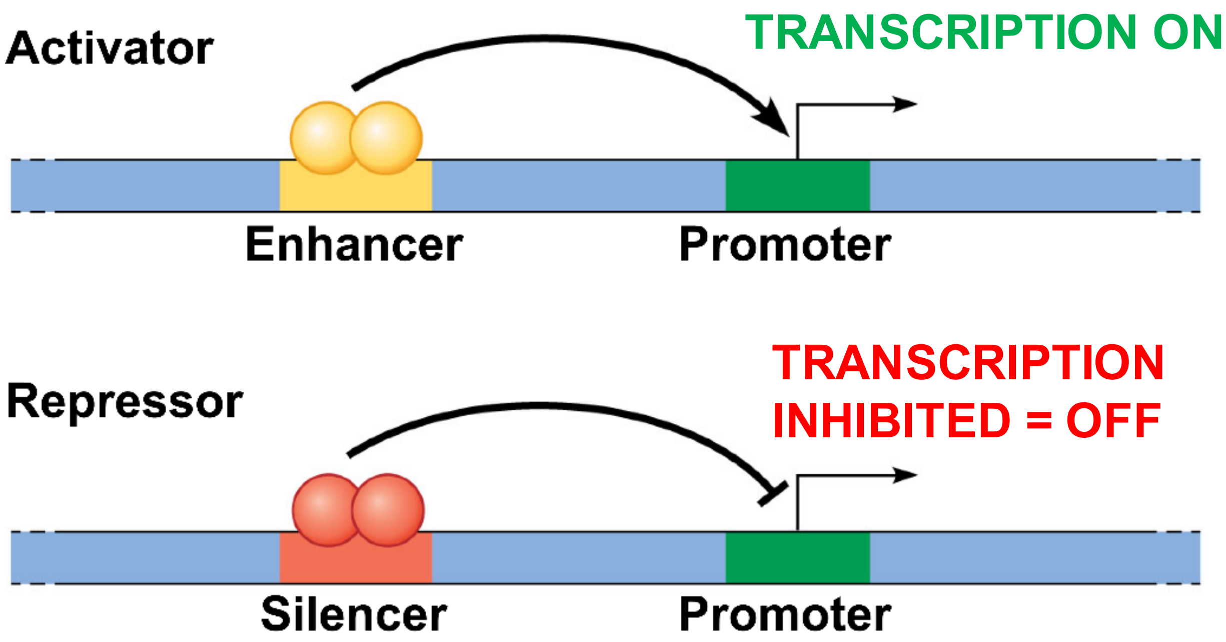
Gene for Albumin, a protein important for wound healing/blood transport

In the absence of activators or inhibitors, transcription can still happen for many genes

2. Transcription factors: proteins that influence whether or not transcription starts

ICA Q5: Gene X requires an activator to be expressed. A mutation prevents this activator from binding to DNA. What happens to Gene X expression?

A) Increases
B) Decreases or turns OFF
C) Stays the same
D) Gene X is deleted



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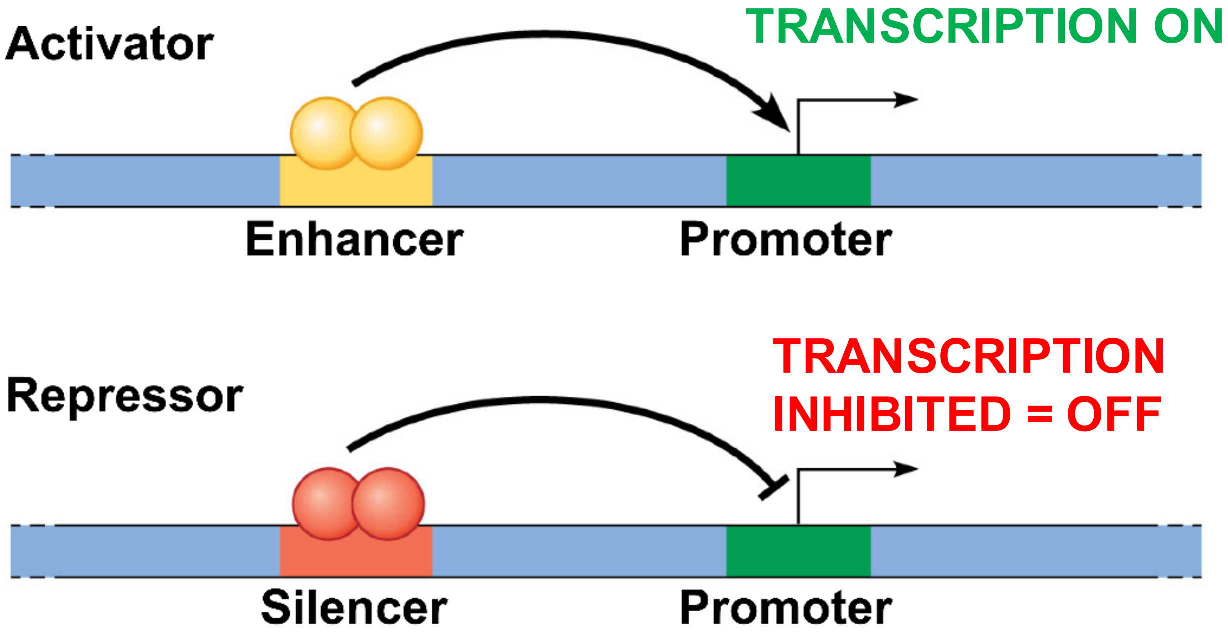


Combinatorial Control of Gene Activation

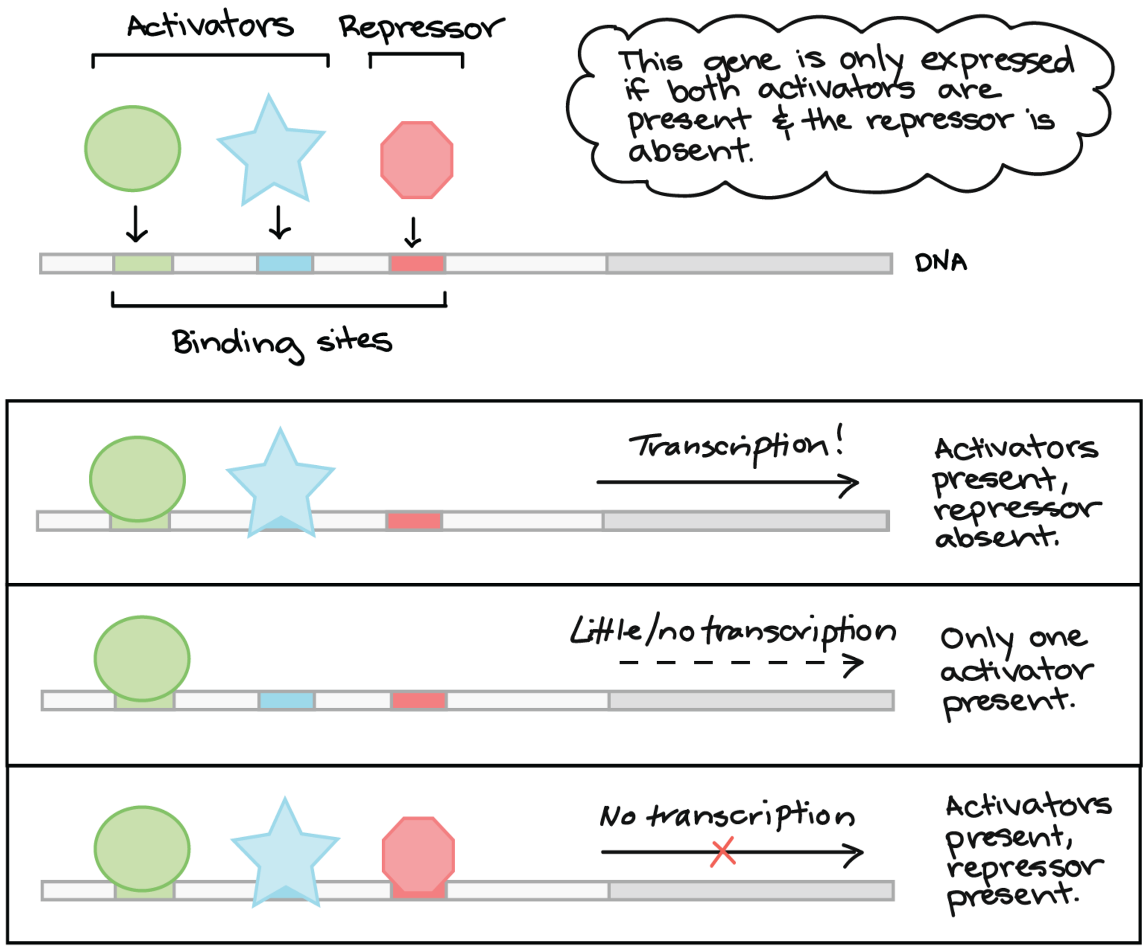
Most genes require MULTIPLE transcription factors. This is called **combinatorial control.**

Different combinations of factors = different outcomes

<u>Example 1</u> for a gene:
- No factors present = OFF
- Only Activator A present = low expression
- Only Activator B present = low expression
- Both A and B present = HIGH expression

<u>Example 2,</u> a gene might need both:
- "Nutrient is available" signal AND
- "Growth factor present" signal
- Only expresses when both conditions are met

Allows the same gene to be regulated differently in different cells



Combinatorial Control of Gene Activation

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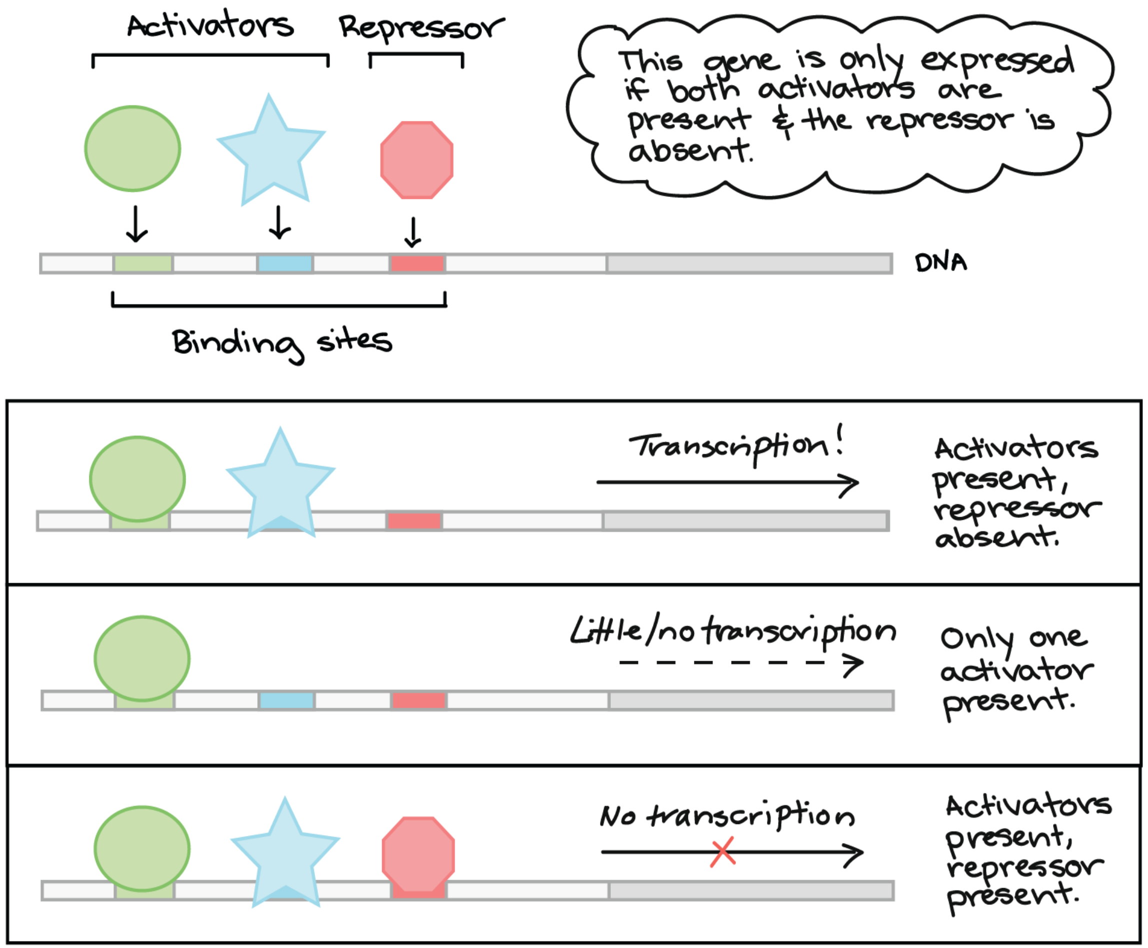
ICA Q6: *Gene Y requires both Activator A and Activator B to be expressed. A cell has Activator A but not Activator B. What happens?*

A) Gene Y is fully expressed
B) Gene Y is not expressed or poorly expressed
C) Gene Y is moderately expressed
D) Cannot determine without more information



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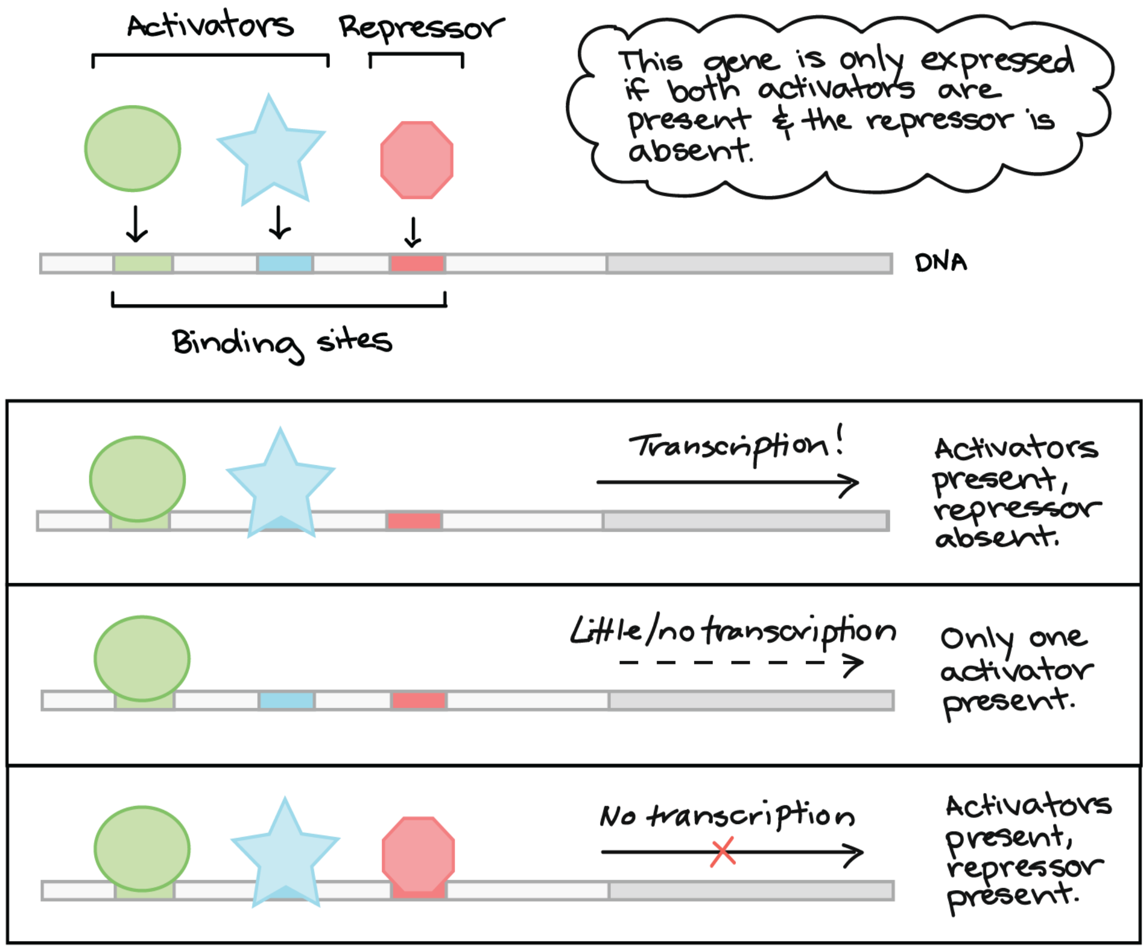
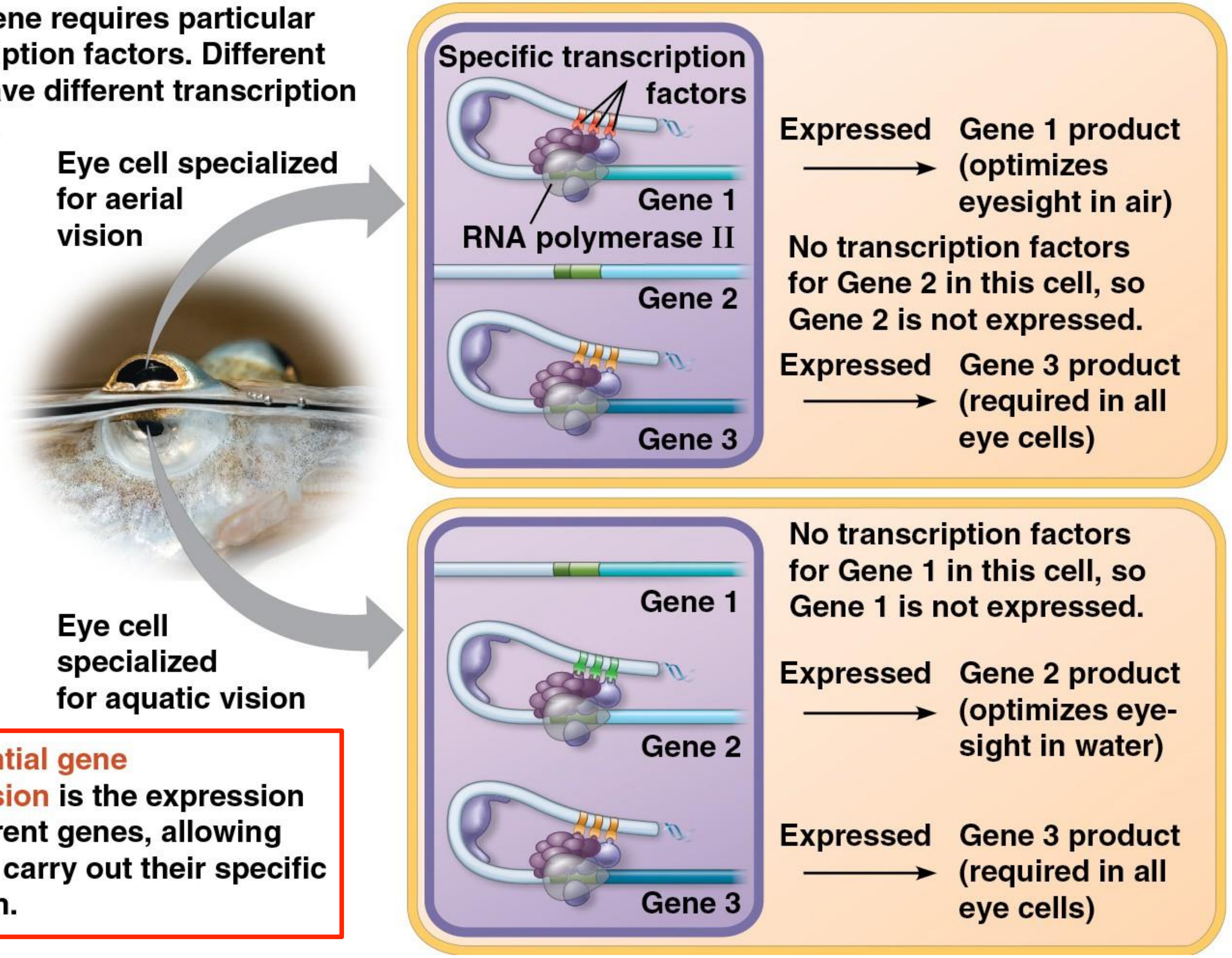


“Four-eyed” fish have eyes raised above the top of the head and divided in two different parts, so that they can see below and above the water surface at the same time. How??

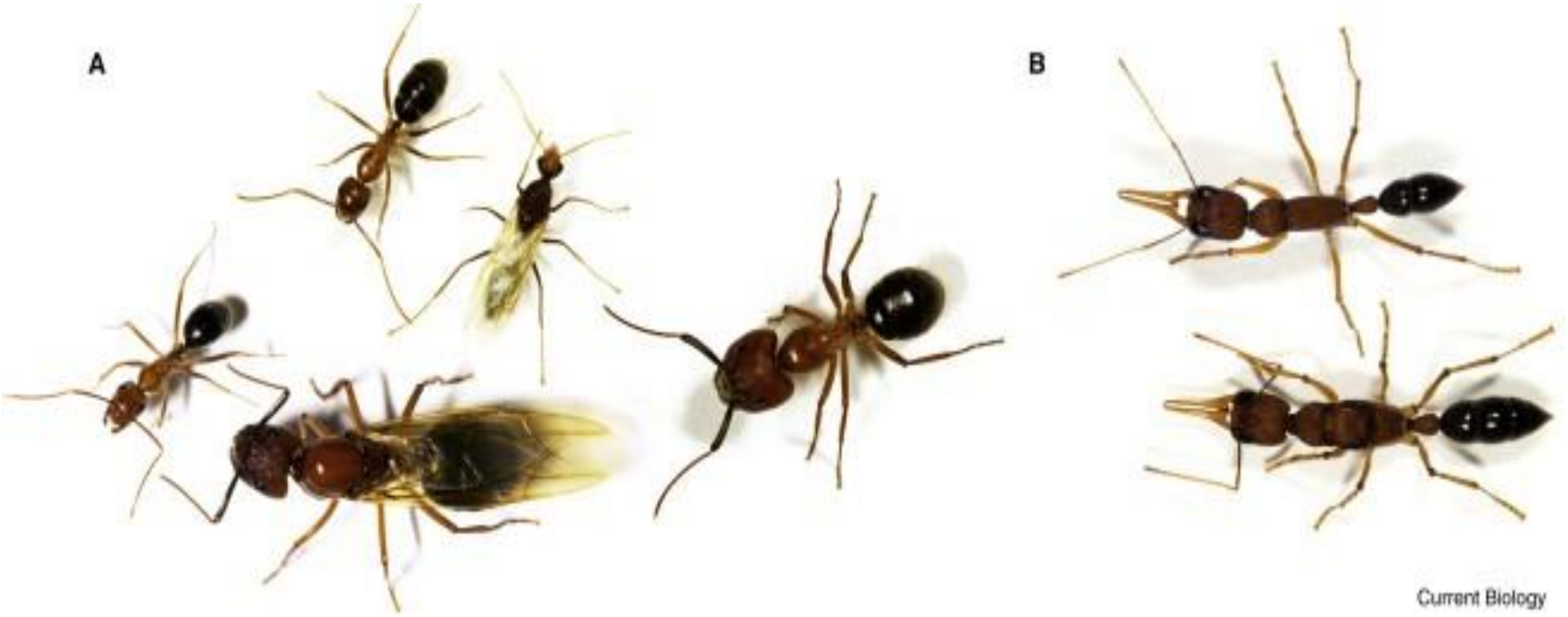
How can two cells with the same set of genes function differently?

Each gene requires particular transcription factors. Different cells have different transcription factors.



Differential gene expression is the expression of different genes, allowing cells to carry out their specific function.

3. Epigenetics: any process that alters gene activity without changing the DNA sequence



ICA Q7: Select your prediction about these two groups of ants:

A. Ants in group A are members of the same species; Ants in group B are members of the same species
B. Ants in group A are members of the different species; Ants in group B are members of the same species
C. Ants in group A are members of the same species; Ants in group B are members of the different species
D. Ants in group A are members of the different species; Ants in group B are members of the different species

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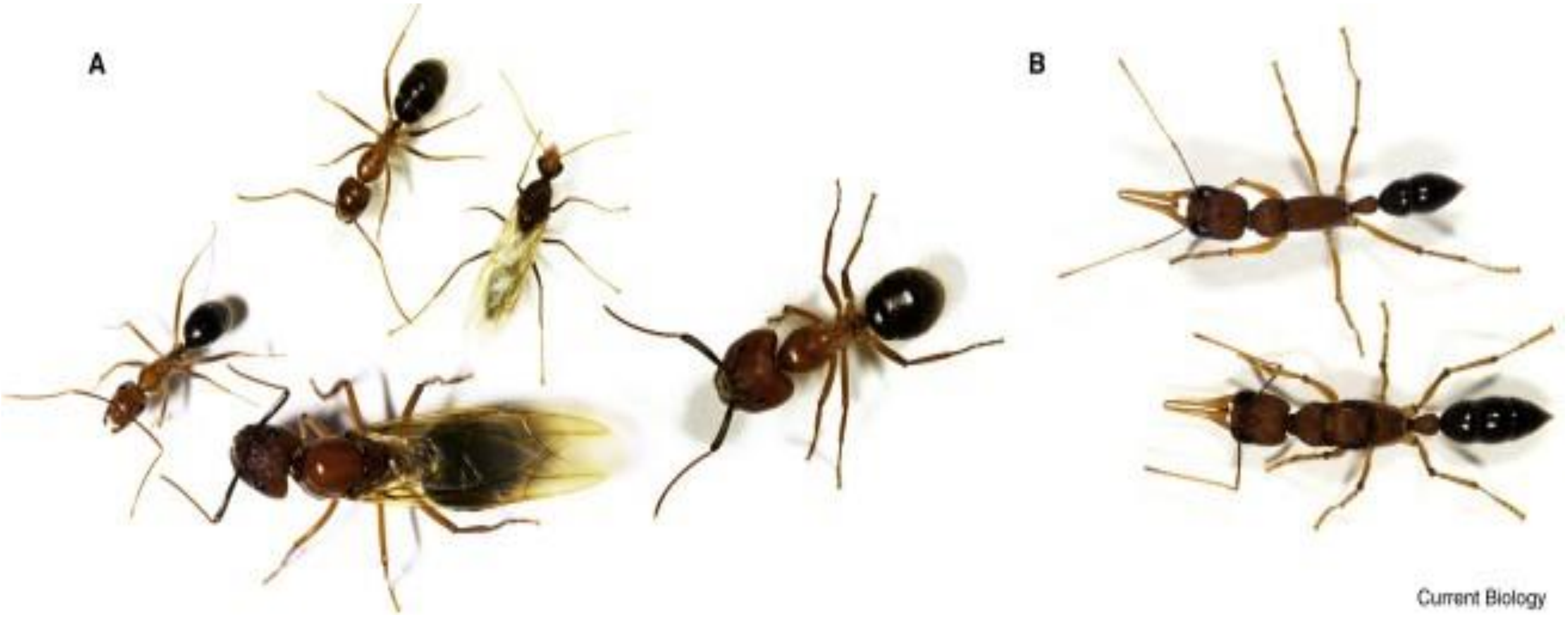

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C. Ants in group A are members of the same species; Ants in group B are members of the different species

D. Ants in group A are members of the different species; Ants in group B are members of the different species

Epigenetics can drive diversity of phenotypes within species without ever changing a gene!

Camponotus floridanus

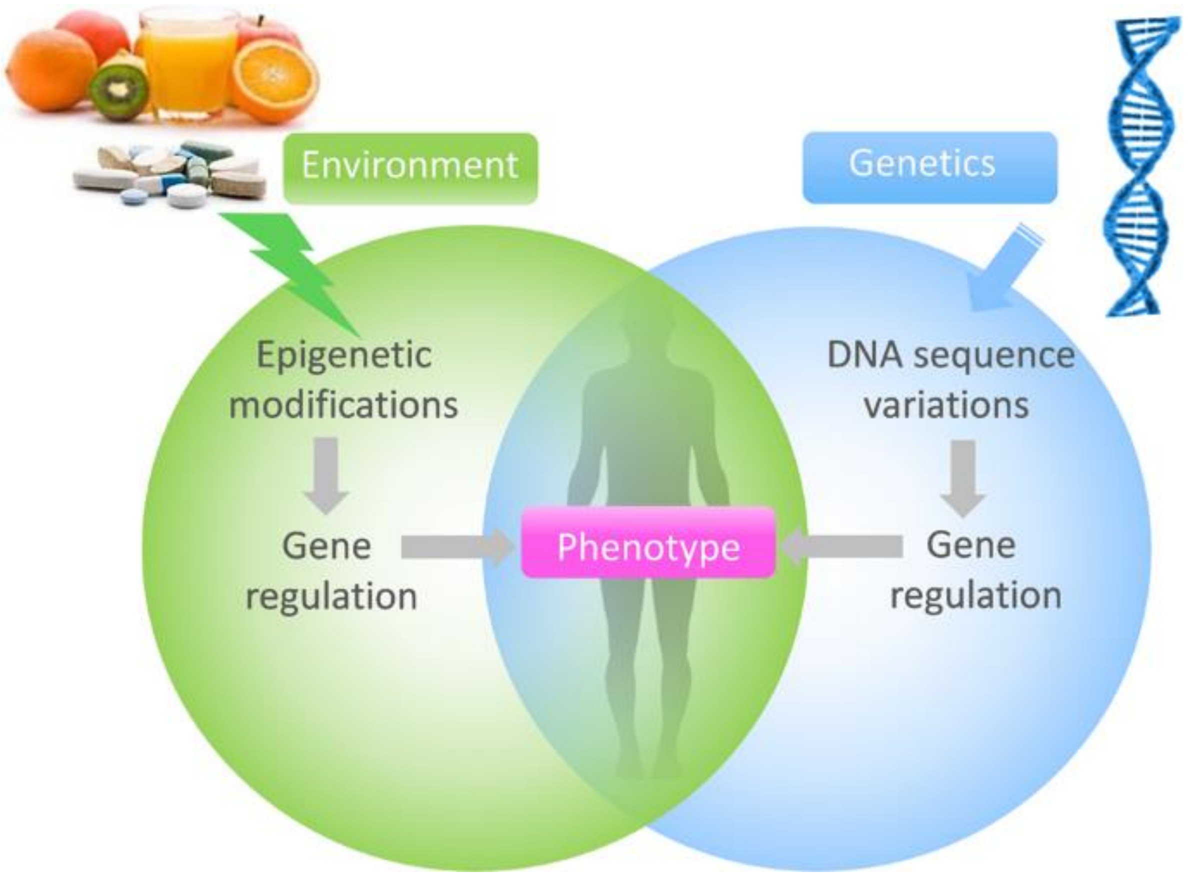
A female embryo can become any one of these ants!

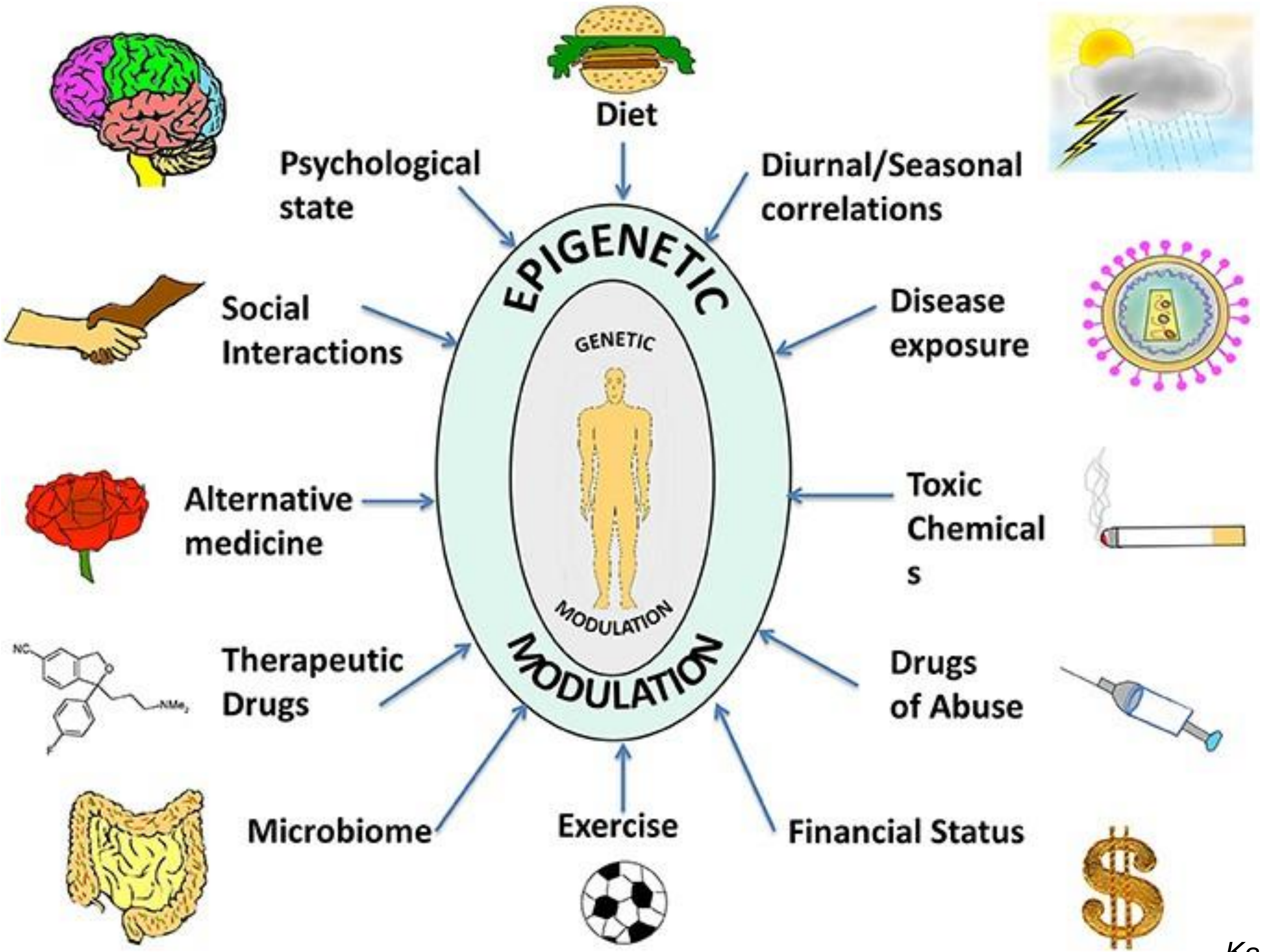
Epigenetics

Greek prefix *epi-* means over, outside of, around

Features that are "on top of" or "in addition to" the traditional genetic basis for inheritance

Any process that alters gene activity without changing the DNA sequence



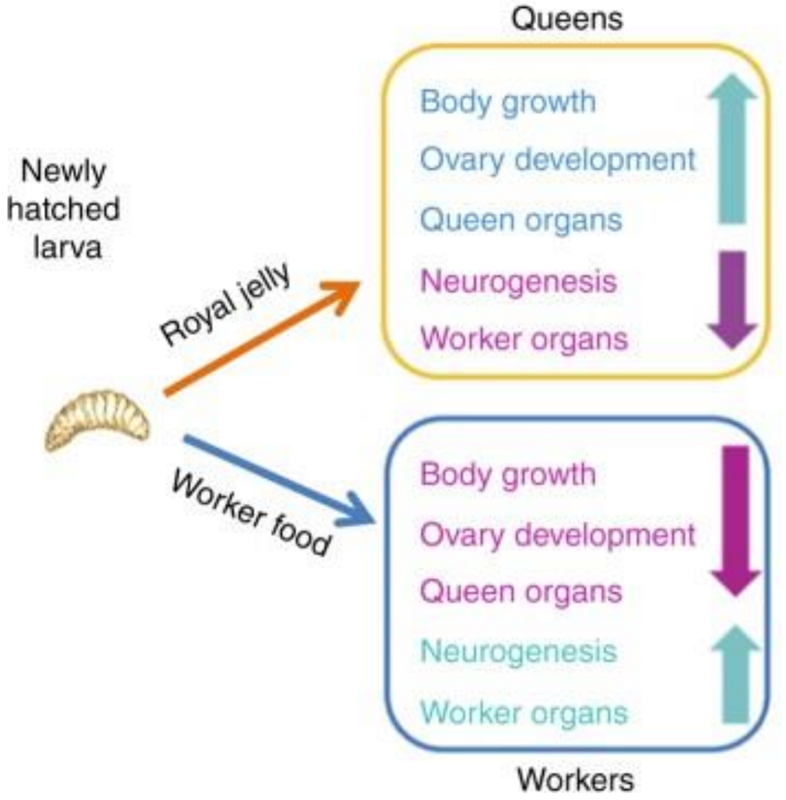


Kanherkar et al. 2014

Like ants, Honeybee queens also develop as a result of epigenetics

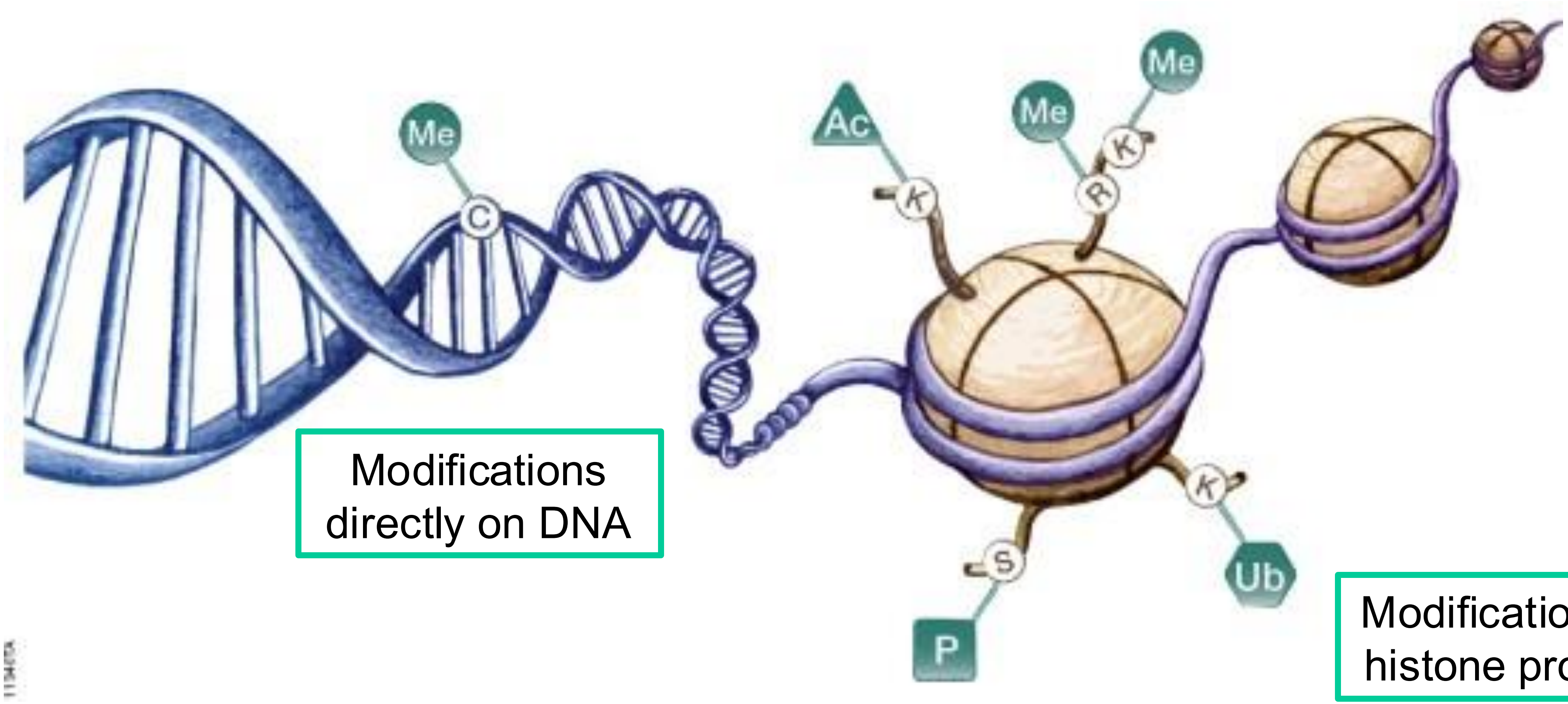
Newly hatched female larvae are multipotent and can develop either into short-lived functionally sterile workers or long-lived fertile queens depending on the feeding regime during larval growth

Queen Bee Larvae: Queens are raised in specially constructed cells called "queen cups," which are filled with royal jelly.



Maleszka R (2018) Commun Biol

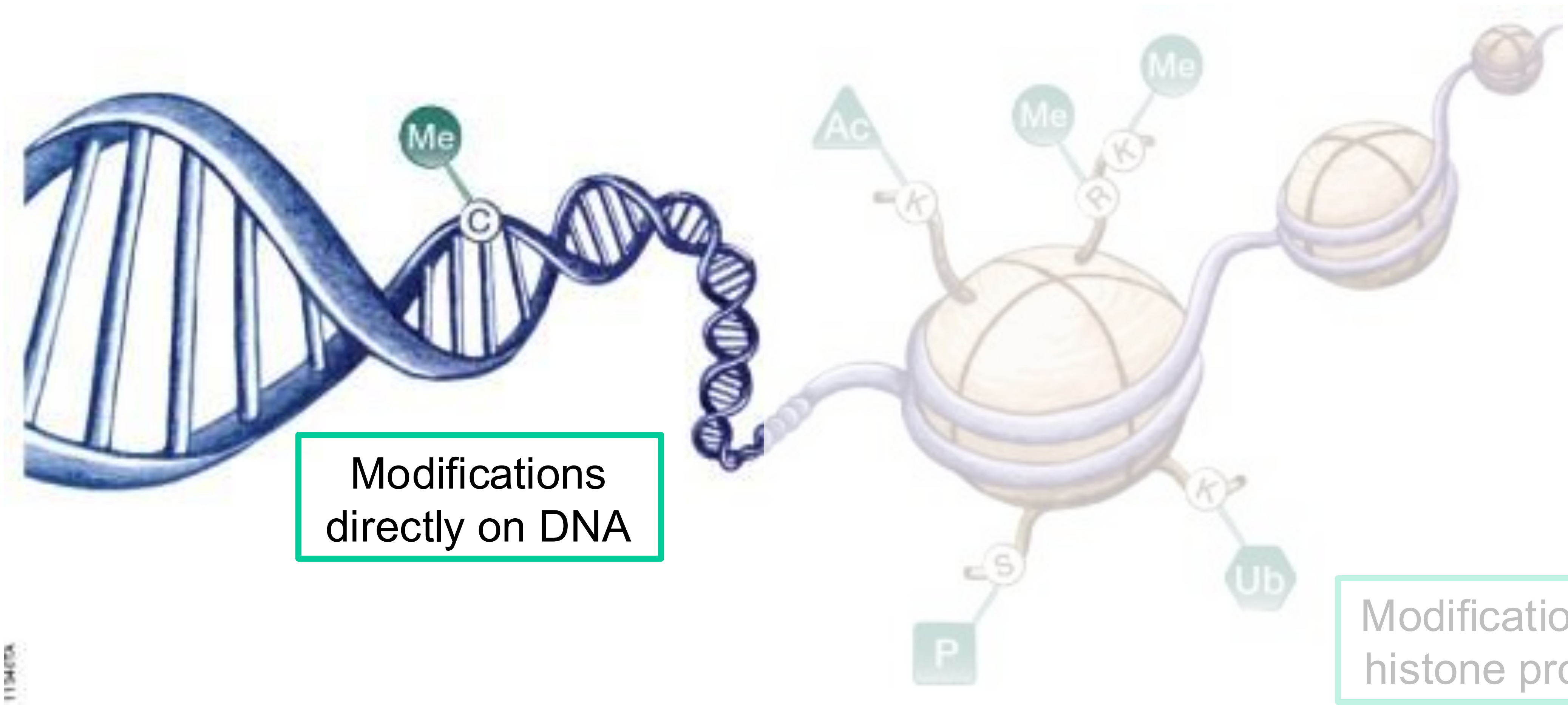
Two ways epigenetics influence gene expression



Modifications directly on DNA

Modifications on histone proteins

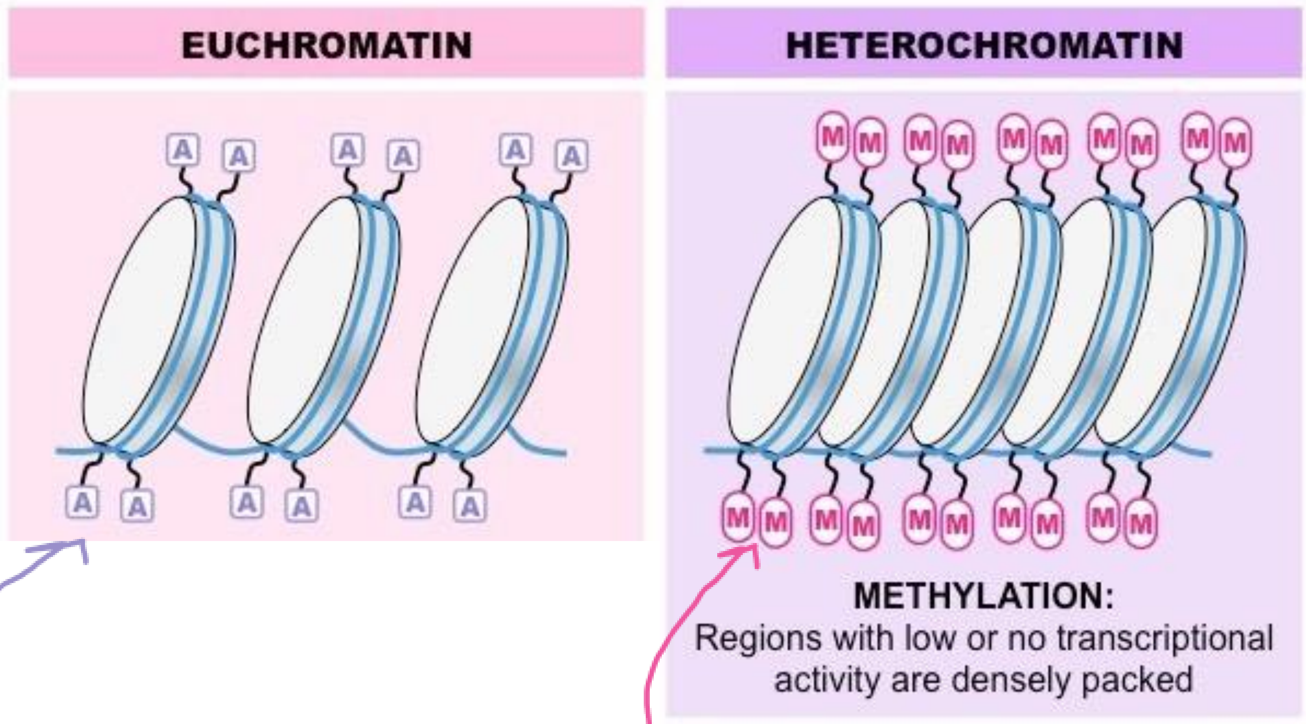
Two ways epigenetics influence gene expression



Modifications directly on DNA

Modifications on histone proteins

DNA Methylation influences open vs closed DNA <u>without changing DNA sequences (the A/G/C/Ts)</u>



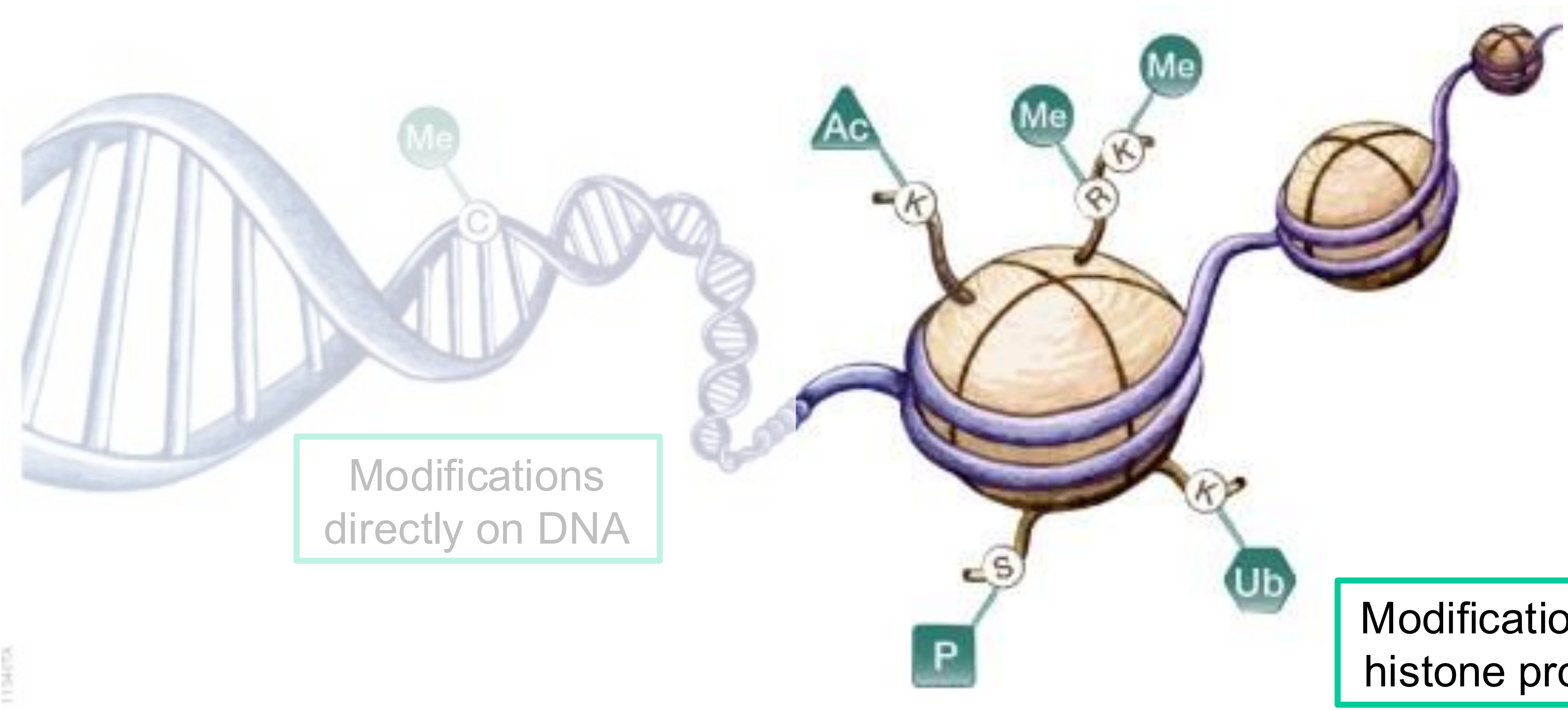
Not methyl groups

Methyl groups added to DNA, but does not change the actual sequence, just an accessory

When DNA is supercoiled and not accessible for transcription, it exists as condensed **heterochromatin**

When the DNA is loosely packed and therefore accessible to the transcription machinery, it exists as **euchromatin**

Two ways epigenetics influence gene expression

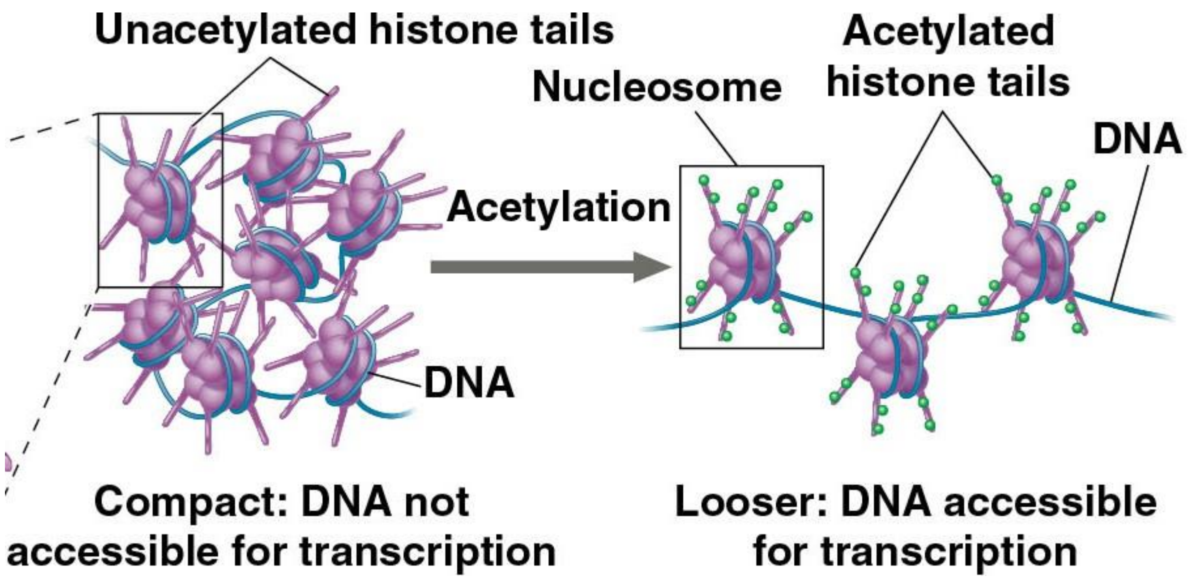


Modifications directly on DNA

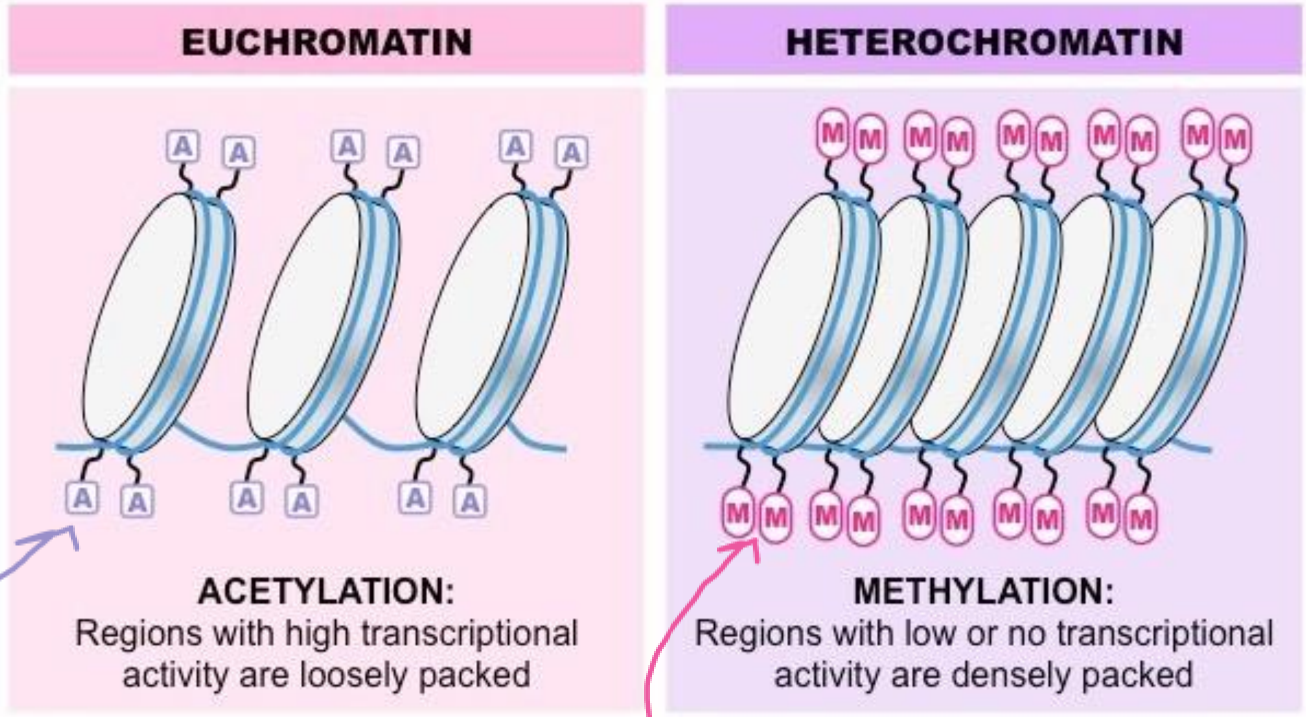
Modifications on histone proteins

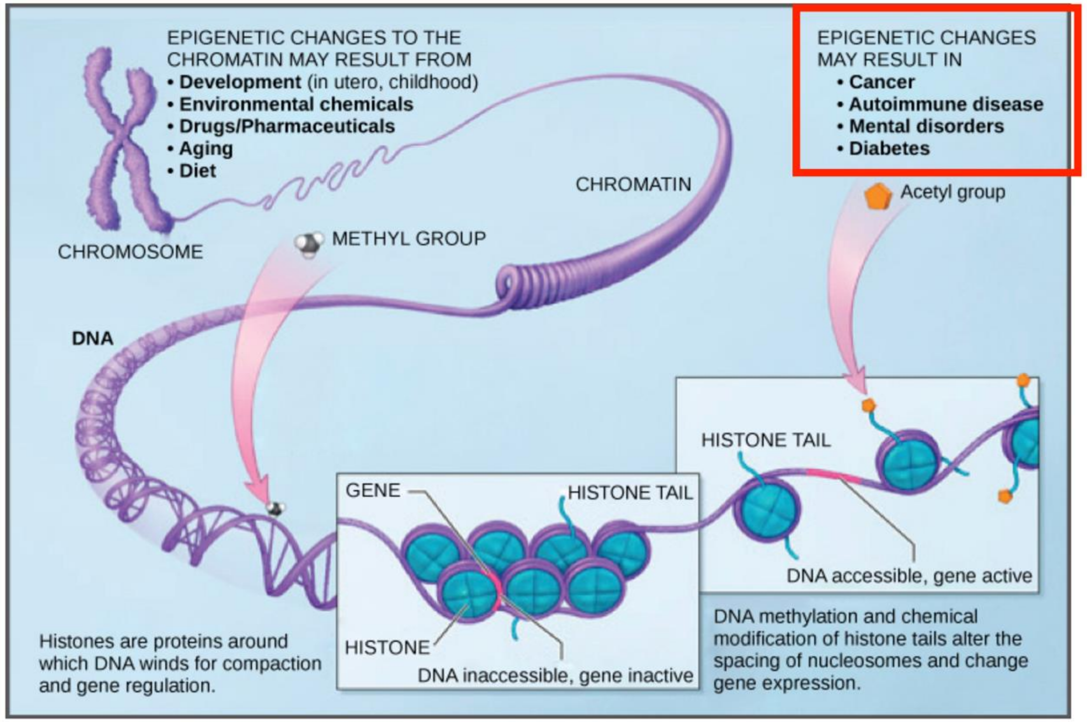
An example of a histone modification…

- In **histone acetylation**, acetyl groups are attached to an amino acid in a histone tail
  - Reduces the positive charge of the histone
- This appears to open up the chromatin structure, thereby promoting the initiation of transcription



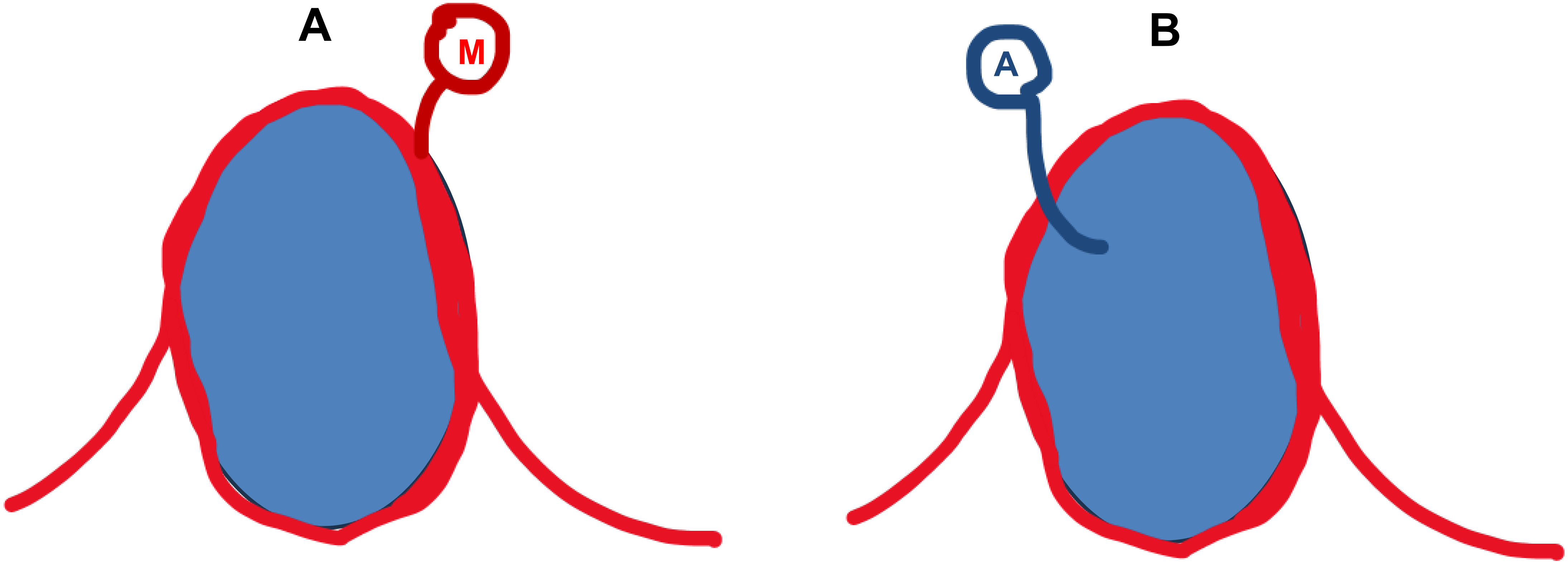
****When gene expression needs to be turned off, **histone deacetylases (HDACs)** remove acetyl groups and DNA becomes tightly wound and inaccessible again*

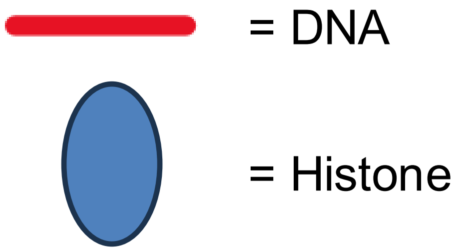




Acetyl groups added to the histones, did not change anything about the DNA

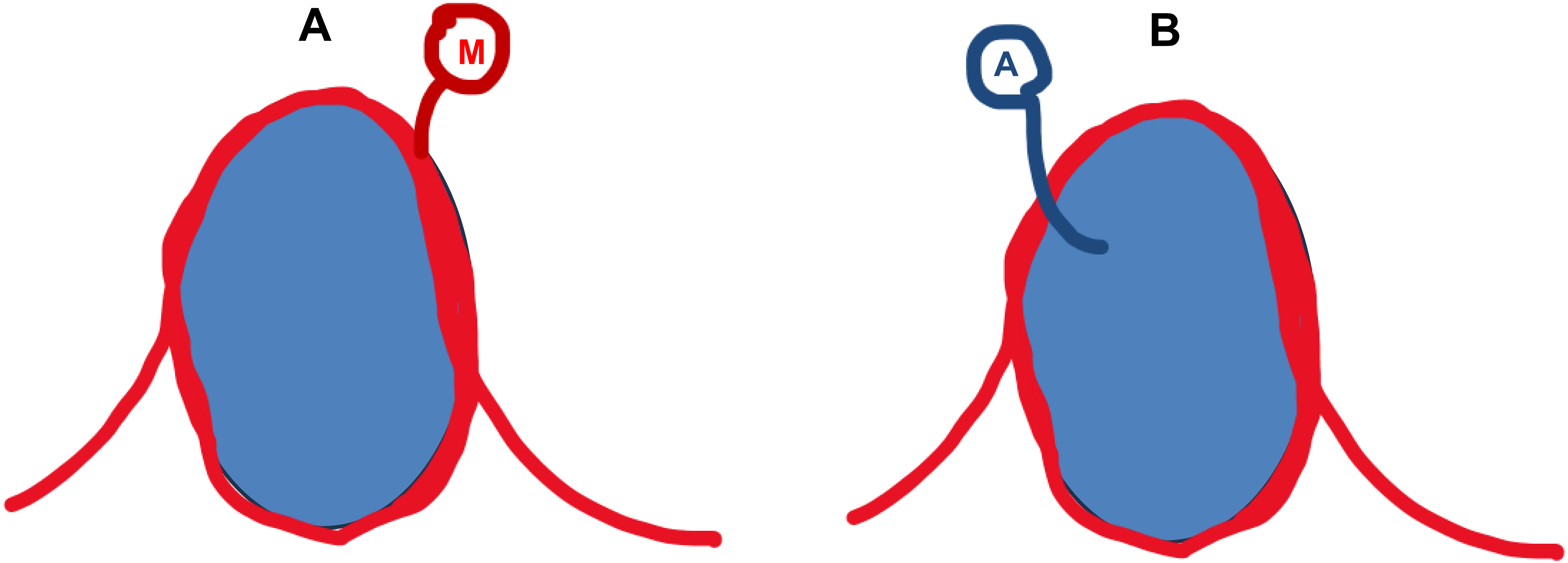
Methyl groups added to DNA, but does not change the actual sequence, just an accessory

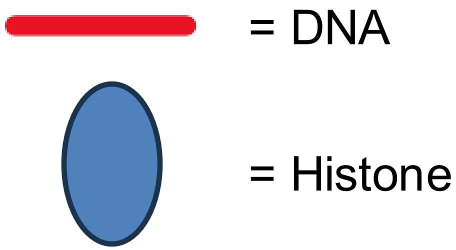




ICA Q8: Each of these pictures has a histone with DNA wrapped around it.

a. Which has a marker on the DNA? A / B
b. Which has a marker on the Histone? A / B
c. Which do you expect to be in regions of open chromatin? A / B
d. Which do you expect to be in regions of closed chromatin? A / B





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Epigenetic Inheritance

- Although the chromatin modifications just discussed do not alter DNA sequence, they may be passed to future generations of cells
- The inheritance of traits transmitted by mechanisms not directly involving the nucleotide sequence is called **epigenetic inheritance**

Example: The Dutch Hunger Winter
Offspring of women in early pregnancy during that particular winter experience adverse health effects as adults: higher rates of obesity, type 2 diabetes, schizophrenia, and mortality over 68

SCIENCE ADVANCES | RESEARCH ARTICLE

HUMAN GENETICS

DNA methylation as a mediator of the association between prenatal adversity and risk factors for metabolic disease in adulthood

Elmar W. Tobi,[1,2] Roderick C. Slieker,[1] René Luijk,[1,3] Koen F. Dekkers,[1] Aryeh D. Stein,[4] Kate M. Xu,[3,5] Biobank-based Integrative Omics Studies Consortium,* P. Eline Slagboom,[1] Erik W. van Zwet,[3] L. H. Lumey,[1,6†] Bastiaan T. Heijmans[1†‡]

Gene Expression Can Be Regulated at Multiple Steps

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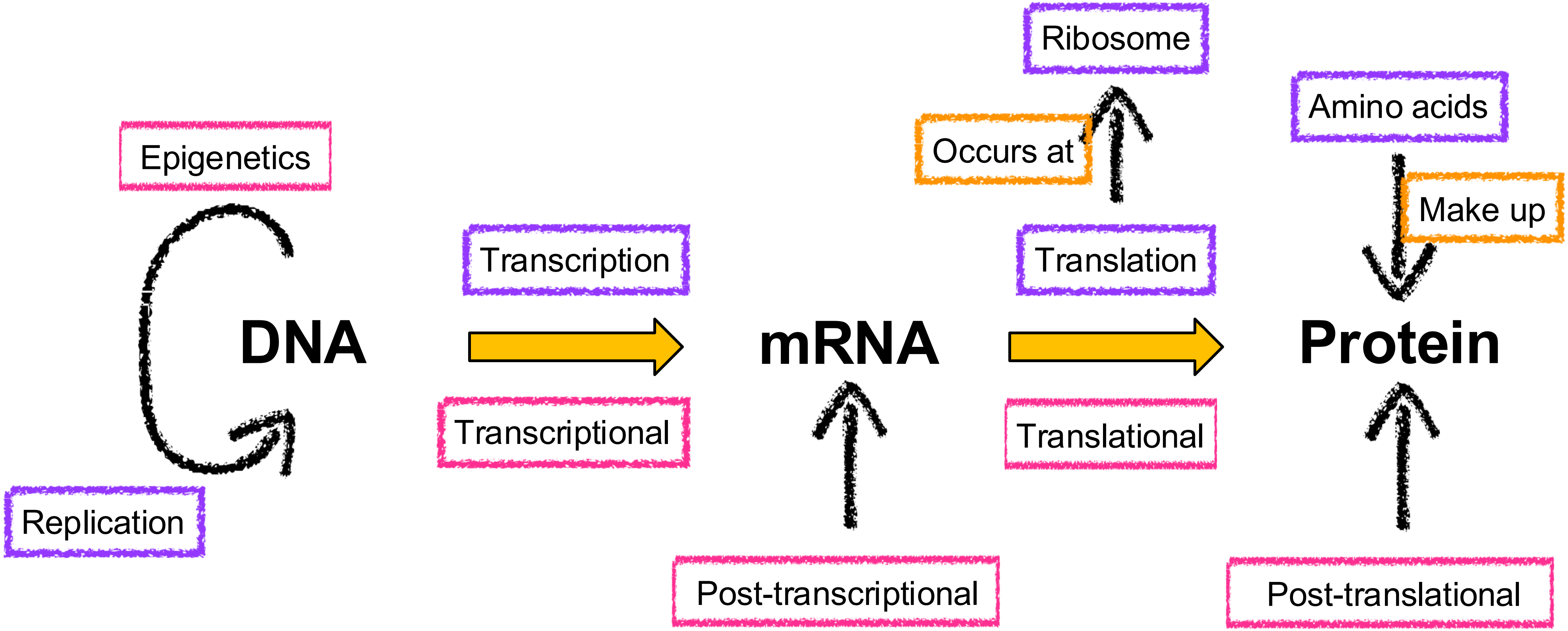
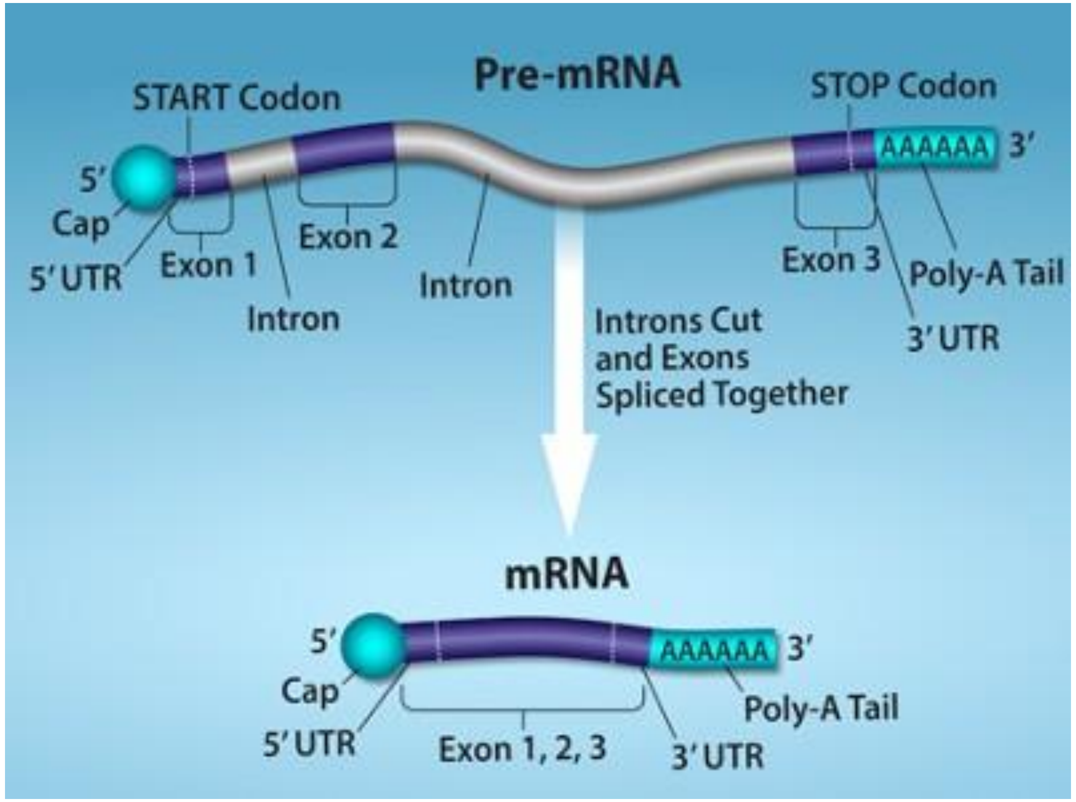
- Translational control (making protein)
- Post-translational control (modifying protein)



Post-Transcriptional Regulation

Regulation Doesn't Stop at Transcription

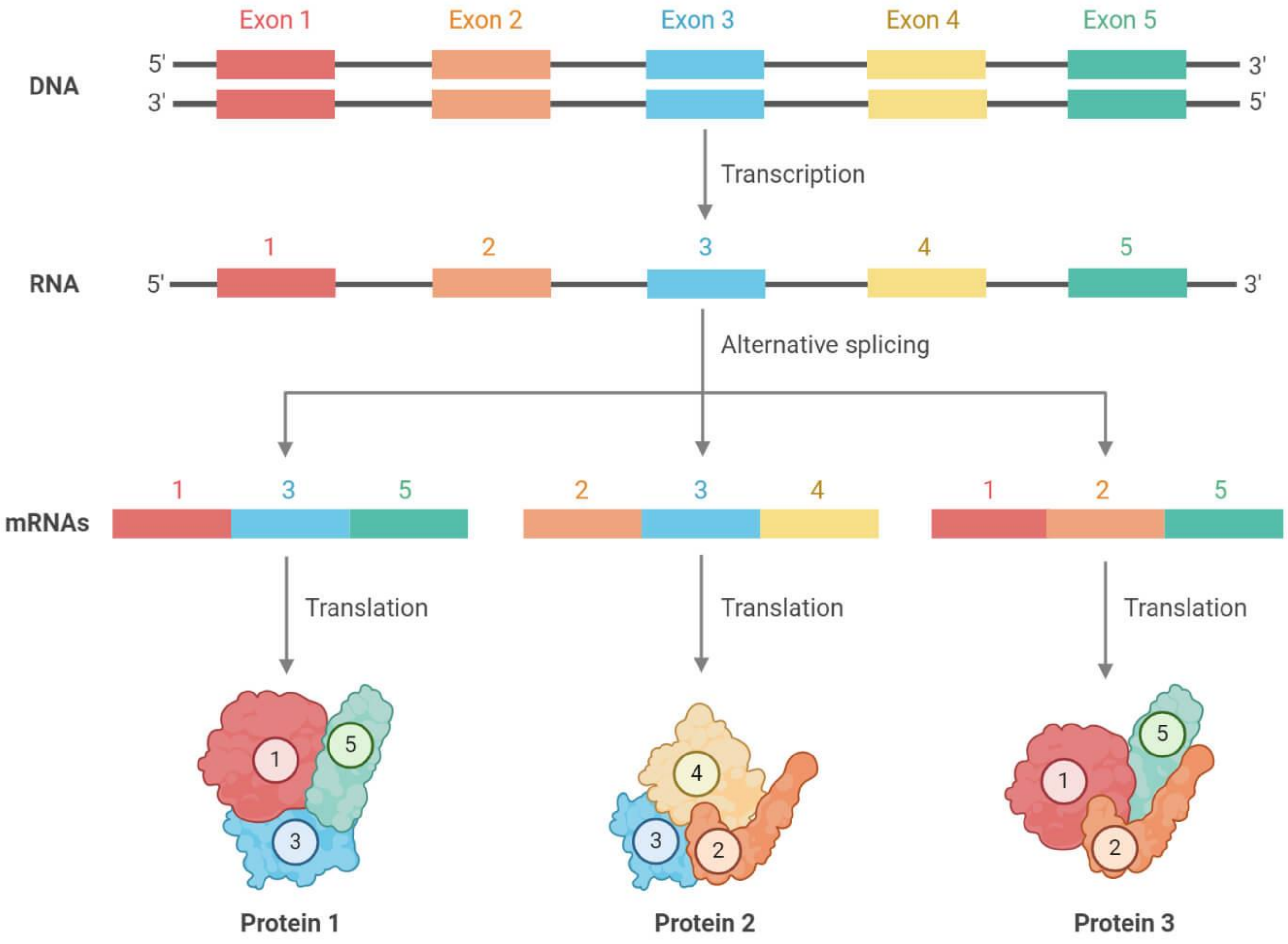
- Once mRNA is made, can still be regulated
- We'll briefly touch on one major mechanism that we have talked about already – splicing!
- This adds flexibility and speed to gene regulation



ALTERNATIVE SPLICING: One gene, many proteins

Example: DSCAM gene (fruit fly)
One gene
Can be spliced 38,016 different ways
Produces 38,016 different proteins
Used in brain development (neuron recognition)

In Humans:
Average gene produces 3-4 different proteins
Allows complexity without huge genome
Different splice variants in different tissues

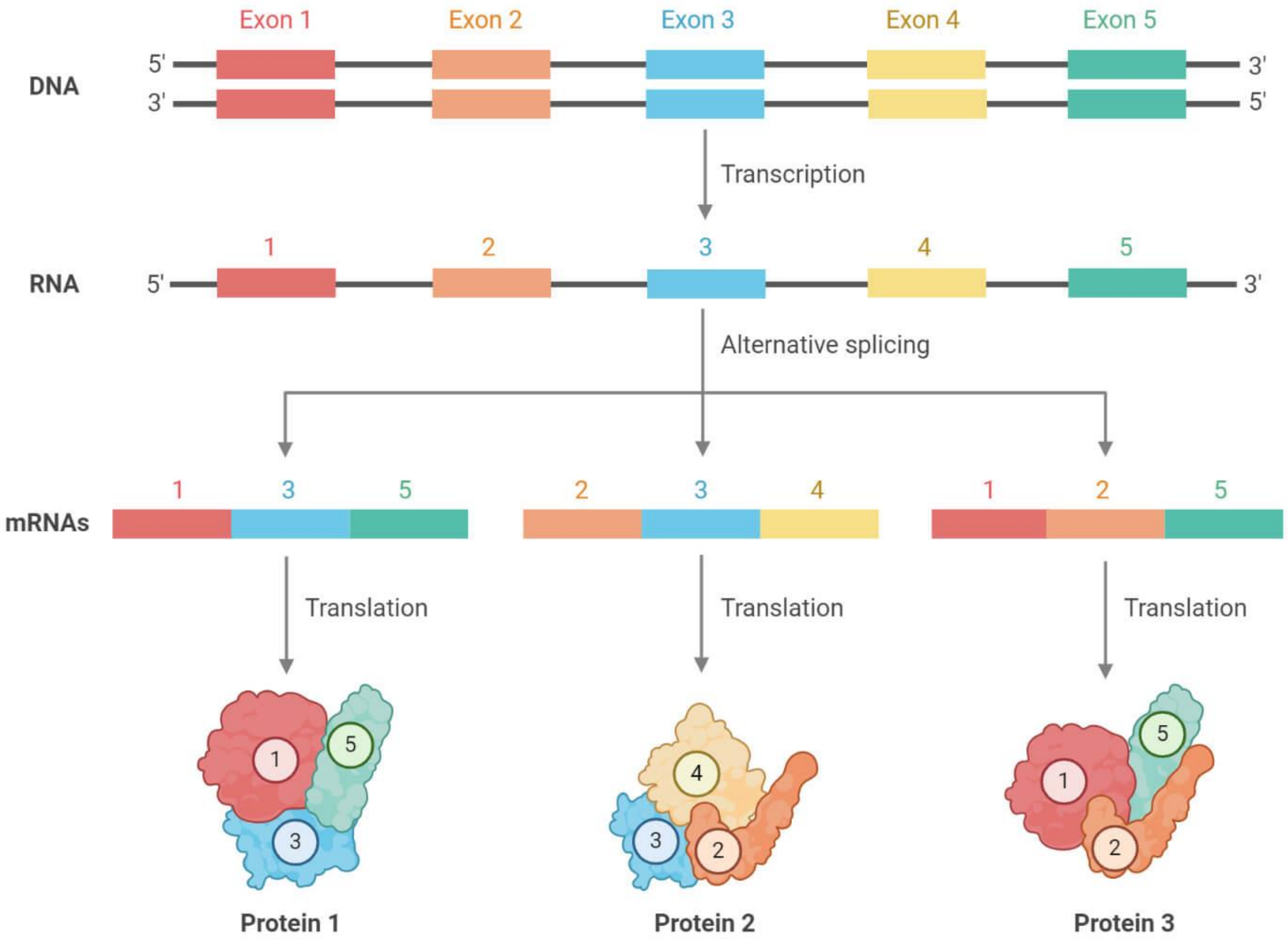


ALTERNATIVE SPLICING: One gene, many proteins

ICA Q9:

Antibody genes produce two forms:
•Membrane-bound version (stays on cell surface)
•Secreted version (released from cell)
•Same gene, different last exon included

Why would this be helpful? How does this show the importance of alternative splicing?

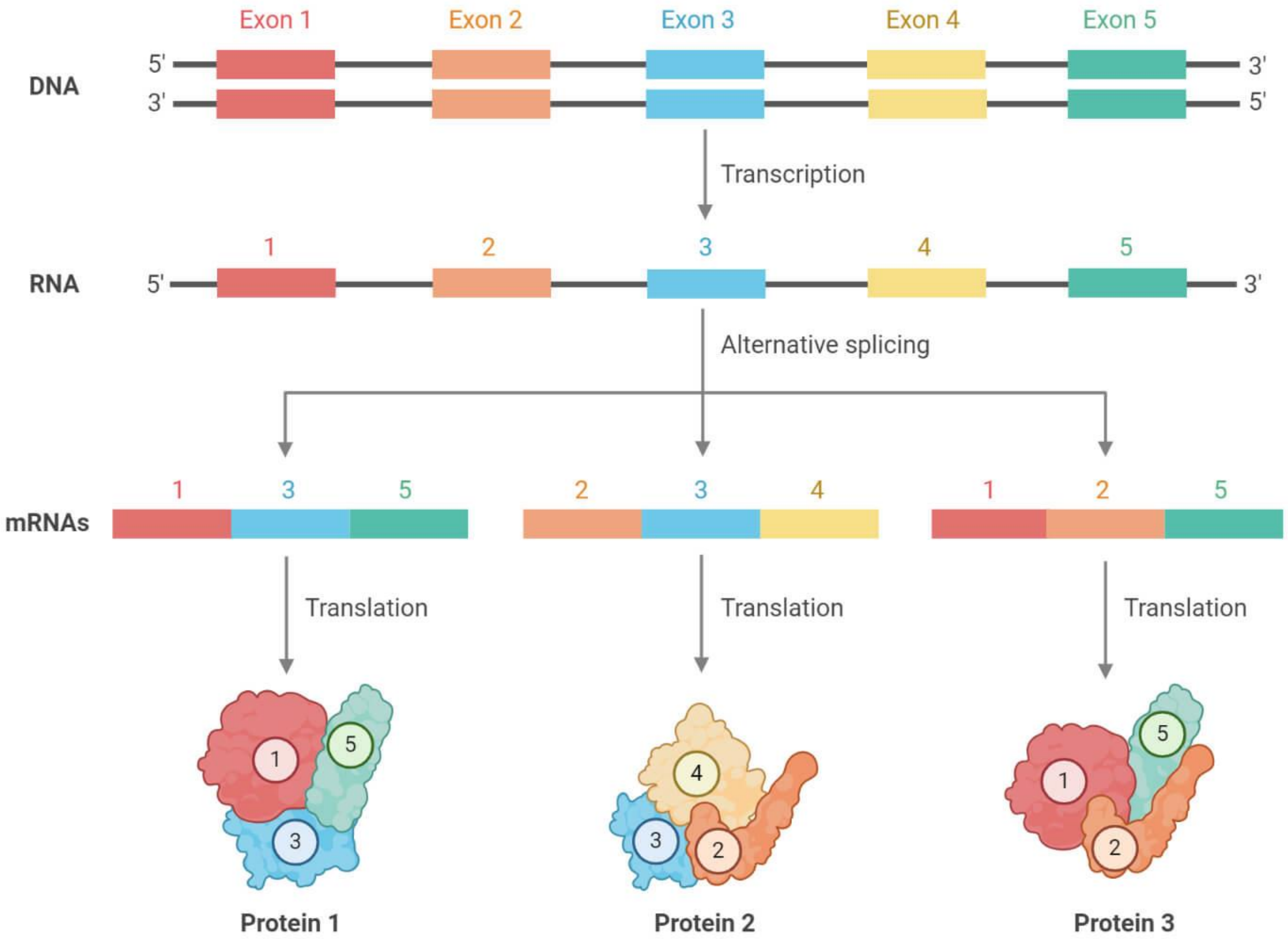


ALTERNATIVE SPLICING: One gene, many proteins

ICA Q9:

Why is making 2 forms of antibodies helpful?
•**Membrane version** = receptor on cell surface to detect germs
•**Secreted version** = released into blood to fight germs

Why does this show alternative splicing is important?
•**One gene does two jobs** without needing two separate genes
•Saves space in the genome
•Cell can quickly switch between forms by just changing splicing
•More efficient!



More than 90% of the human protein-coding genes undergo alternative splicing

- In **alternative RNA splicing**, different mRNA molecules are produced from the same primary transcript
  - Some genes can encode more than one kind of polypeptide, depending on which segments are treated as exons during splicing
- Alternative RNA splicing can significantly expand the repertoire of a eukaryotic genome
- Consequently, the number of different proteins an organism can produce is much greater than its number of genes

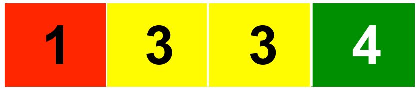
ICA q10: A eukaryotic gene has a pre-mRNA with four exons:

Which of the following arrangements of exons **would** be possible during alternative splicing of a mature mRNA?

A

B

C



D

E

F

A eukaryotic gene has a pre-mRNA with four exons:

1 2 3 4

Which of the following arrangements of exons **would** be possible during alternative splicing of a mature mRNA?

**you can't change the order

A 1 2 3 4

B 1 3 4

~~**C** 1 3 3 4~~

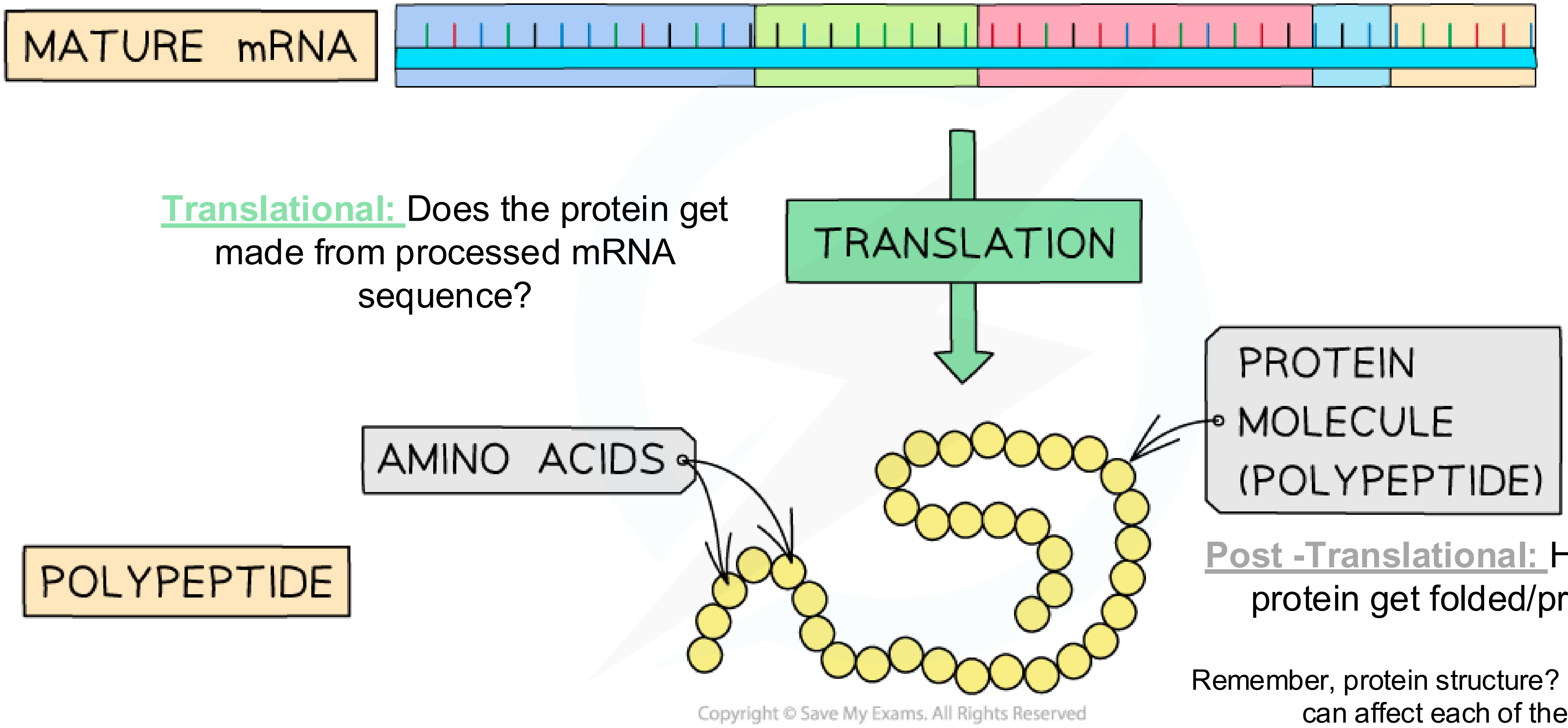
**NO DUPLICATED EXONS

~~**D** 1 3 2 4~~

E 1 2 4

F 1 3

Don’t forget about translational/post-translational control!



Translational: Does the protein get made from processed mRNA sequence?

Post -Translational: How does the protein get folded/processed?

Remember, protein structure? Post-translational can affect each of these steps

- Primary
- Secondary
- Tertiary
- Quaternary

Announcements

- No class on Wednesday
- No lab this week
- Problem set 4B will be uploaded today
- Exam 4 is DURING FINALS WEEK – check the final exam schedule, we follow the MWF schedules.