GENE AND GENOME

Nguyen Le Thanh (MRes)
thanhnl@pnt.edu.vn

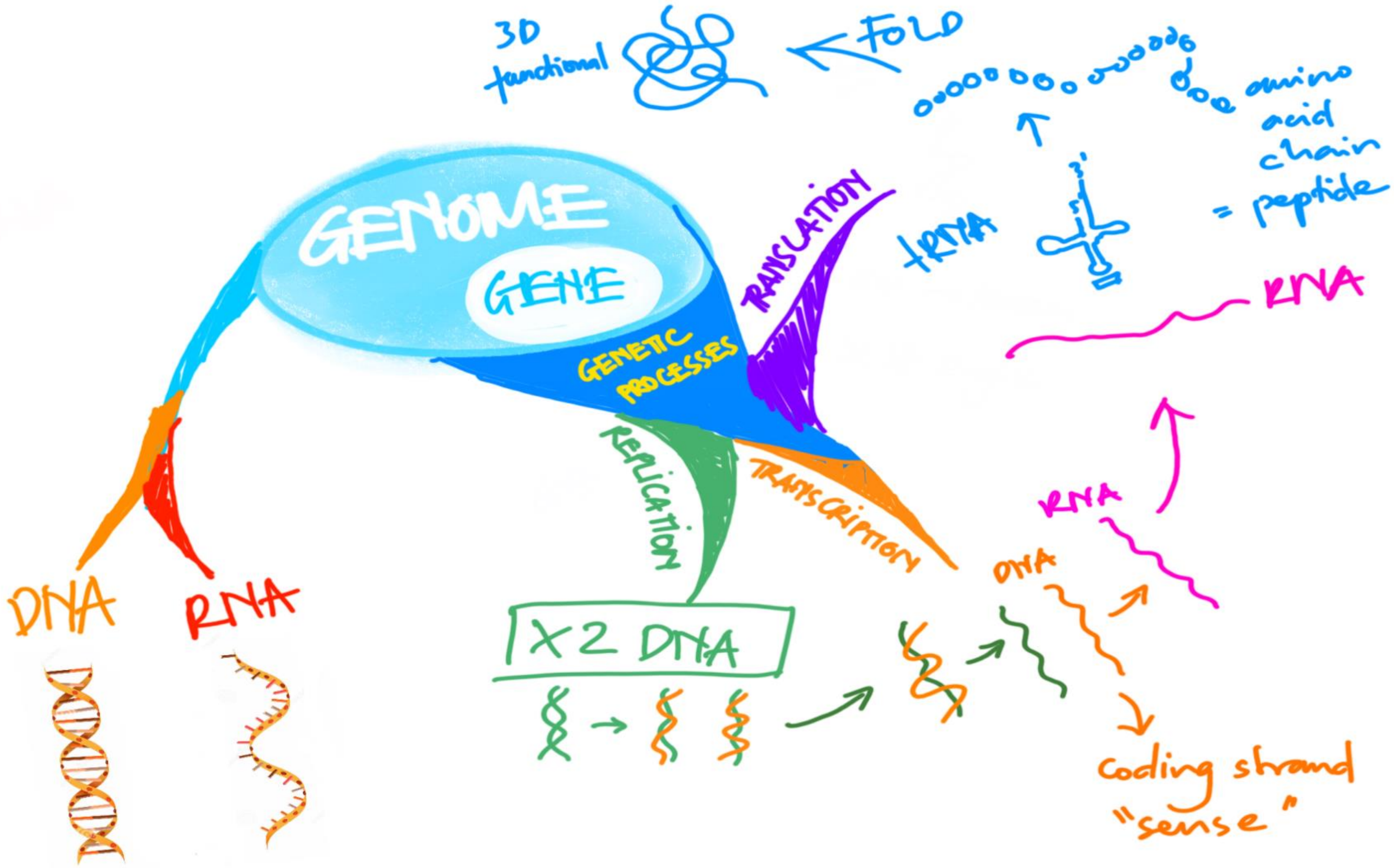
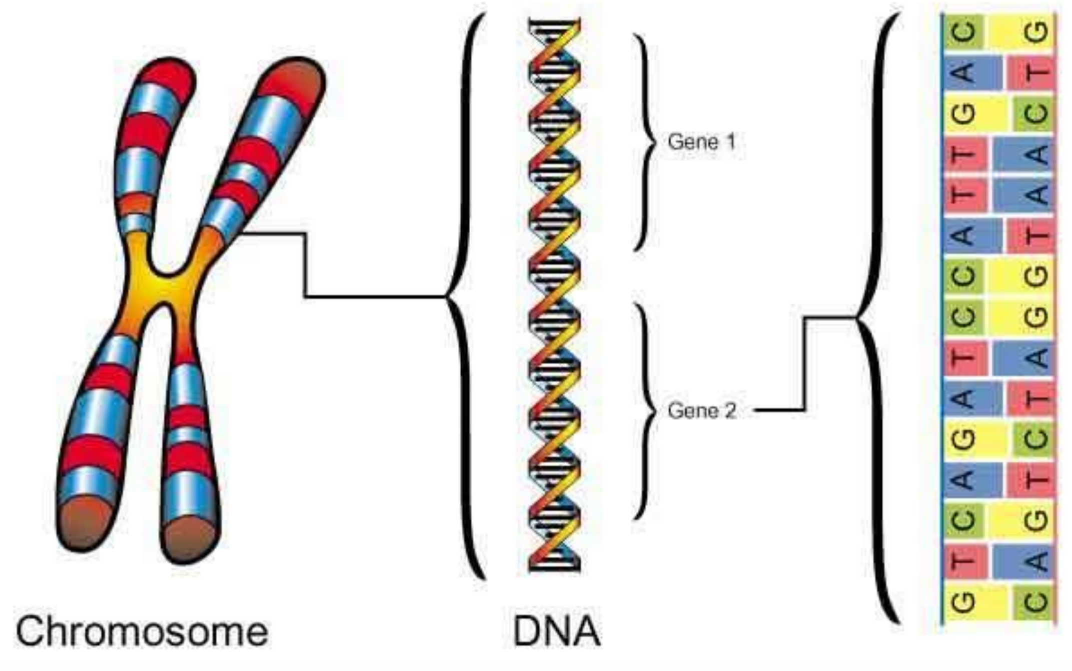
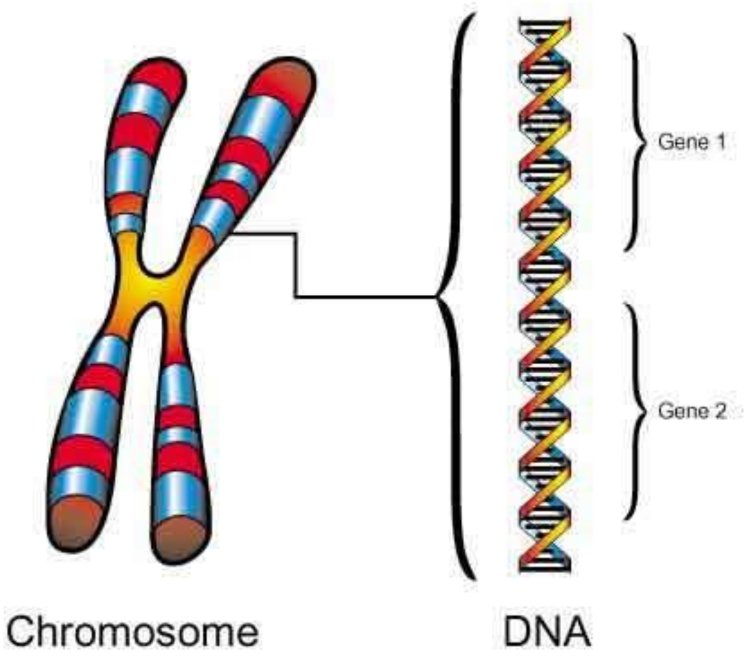

3D functional
FOLD
amino acid chain = peptide
tRNA
RNA
TRANSLATION
GENOME
GENE
GENETIC PROCESSES
REPLICATION
TRANSCRIPTION
DNA
RNA
X2 DNA
RNA
DNA
coding strand "sense"

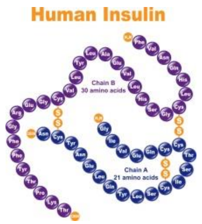
TABLE OF CONTENT

1. GENE AND GENOME

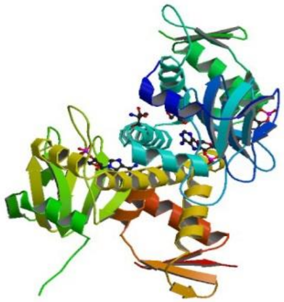

Chromosome
DNA
Gene 1
Gene 2
C G
A T
G C
T A
T A
A T
C G
C G
T A
A T
G C
A T
C G
T A
G C



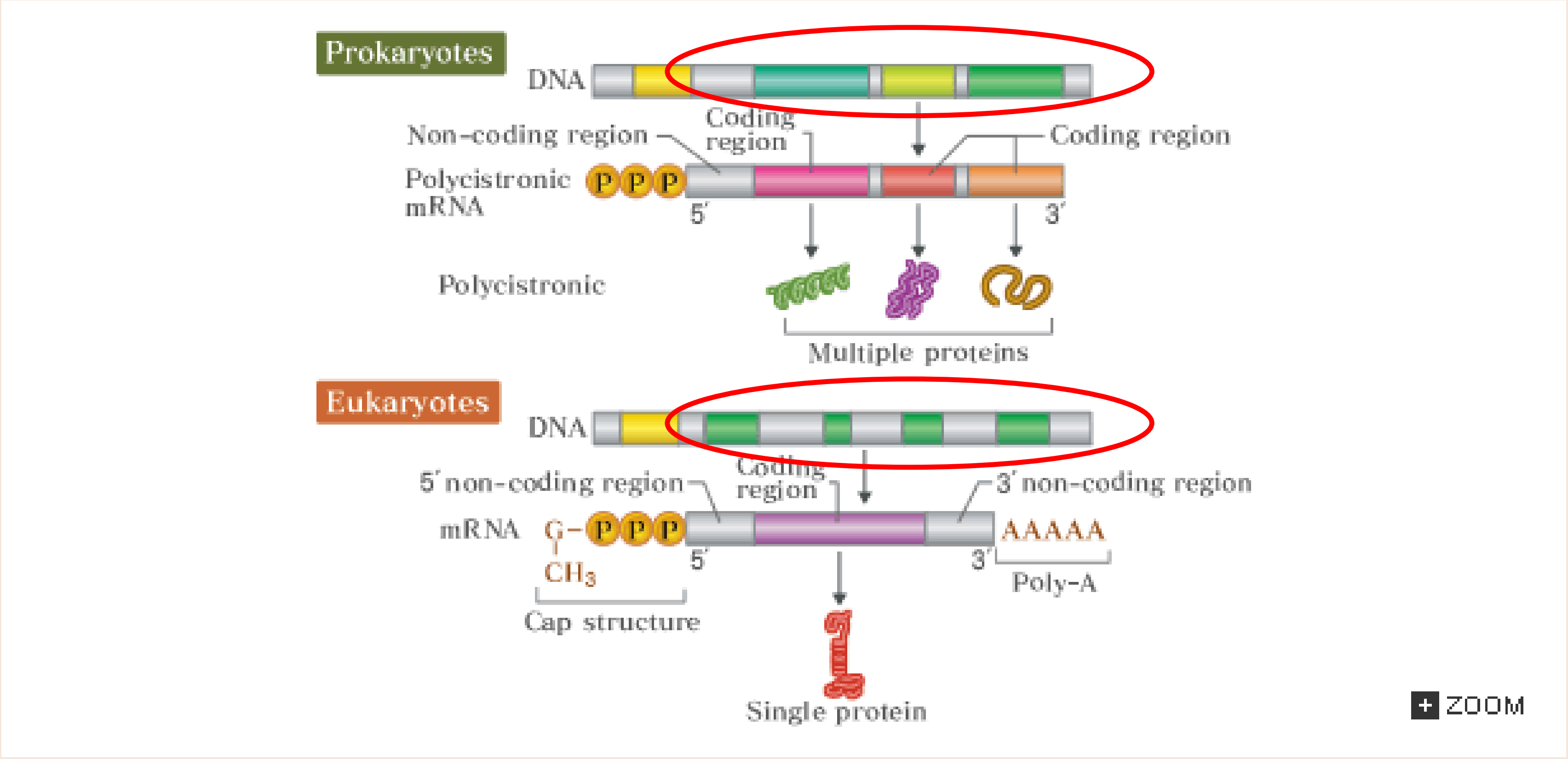
function



regulation



cAMP receptor protein


Prokaryotes
DNA
Non-coding region
Coding region
Coding region
Polycistronic mRNA
P P P
5′
3′
Polycistronic
Multiple proteins
Eukaryotes
DNA
5′ non-coding region
Coding region
3′ non-coding region
mRNA
G–P P P
CH3
5′
3′
AAAAA
Poly-A
Cap structure
Single protein
+ ZOOM


Michelob ULTRA

STRUCTURE OF A GENE

=

A RUNNING TRACK

WARMING AREA

RUNNING TRACK

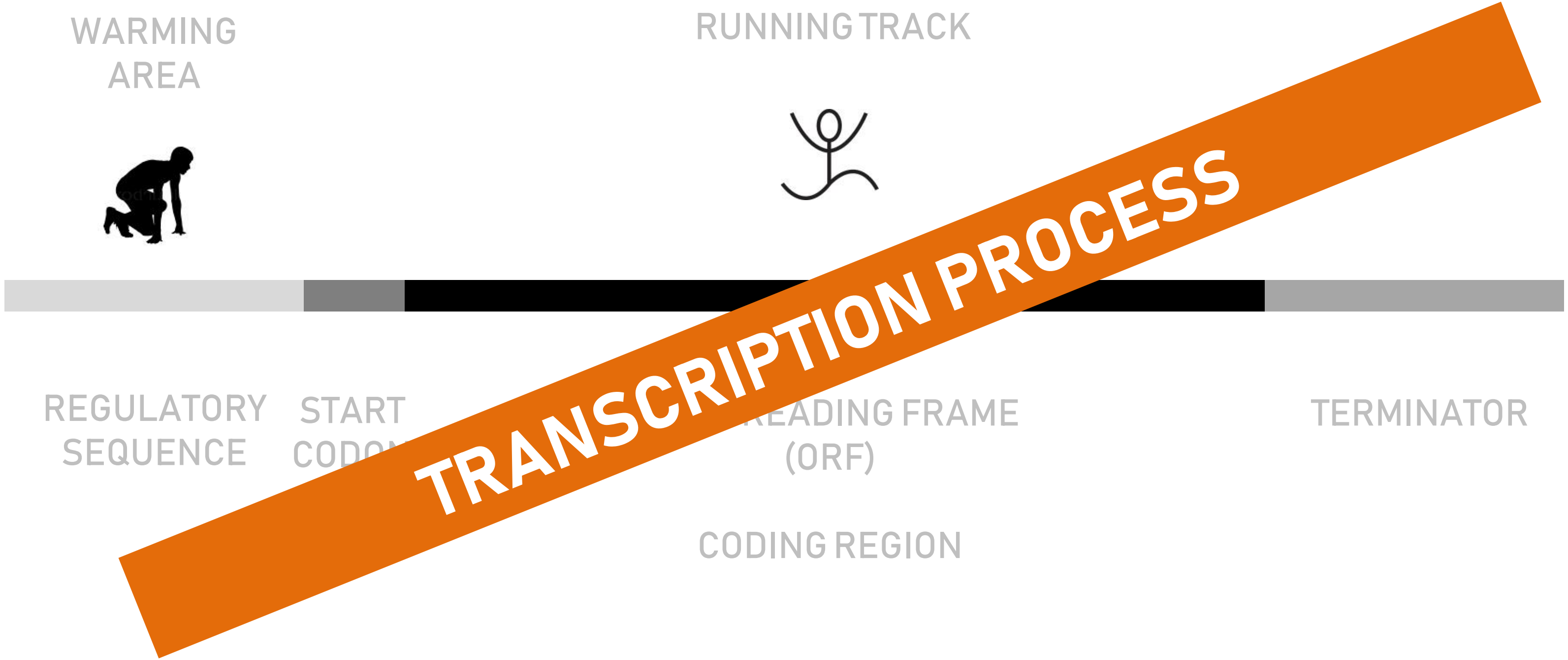
REGULATORY SEQUENCE

START CODON

OPEN READING FRAME (ORF)

TERMINATOR

CODING REGION



WARMING AREA

RUNNING TRACK

REGULATORY SEQUENCE

START CODO

READING FRAME (ORF)

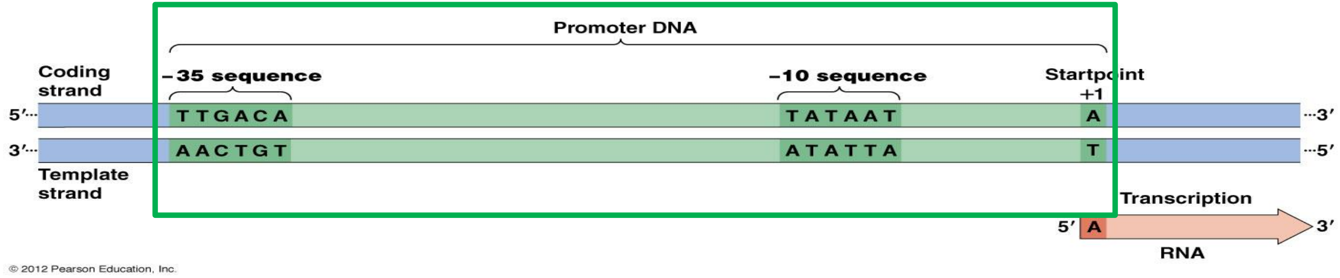
TERMINATOR

CODING REGION

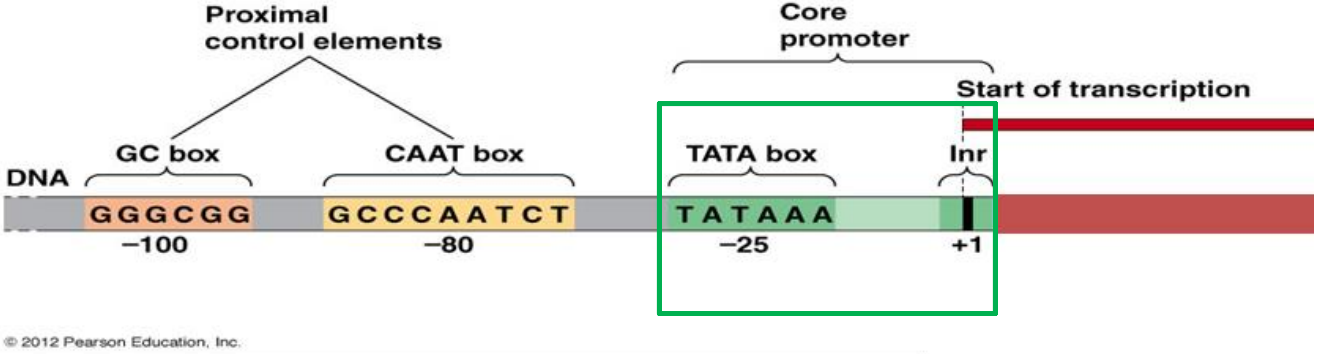
TRANSCRIPTION PROCESS

REGULATORY SEQUENCE

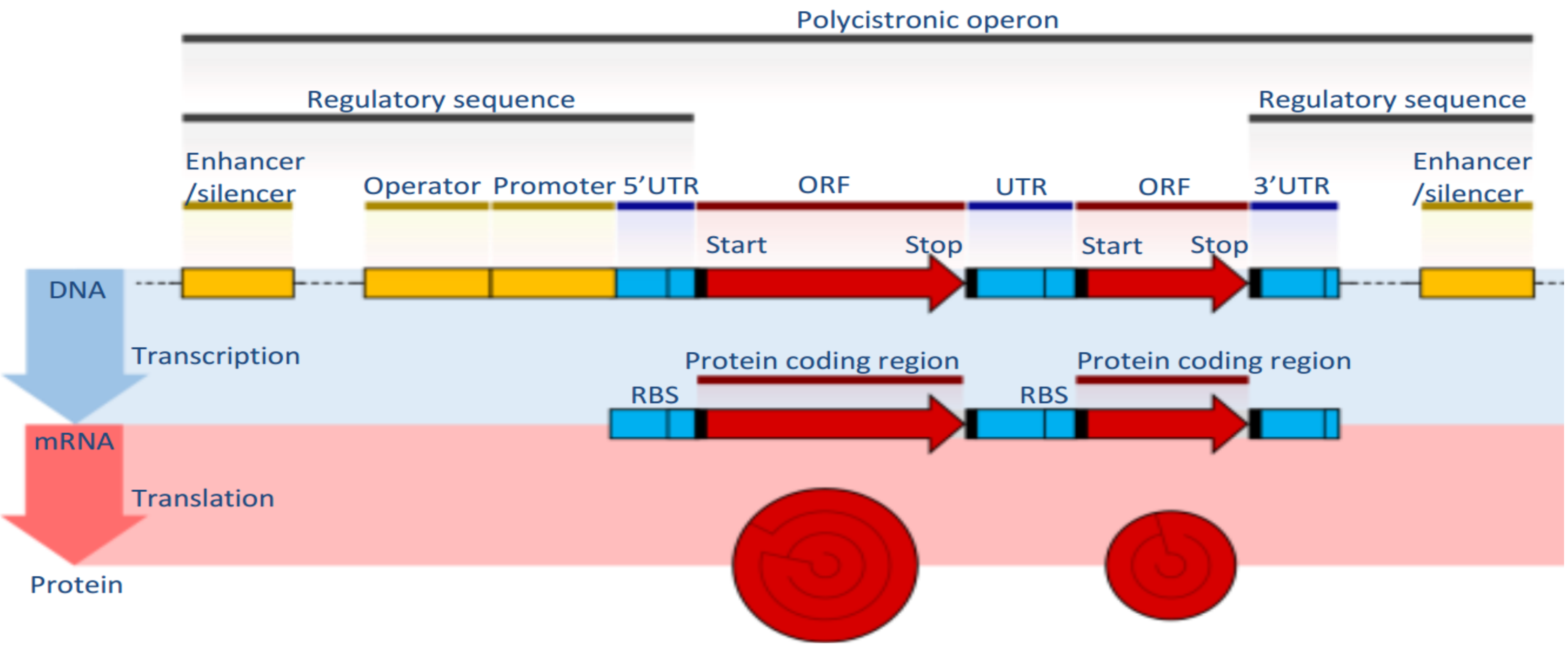
Prokaryotes



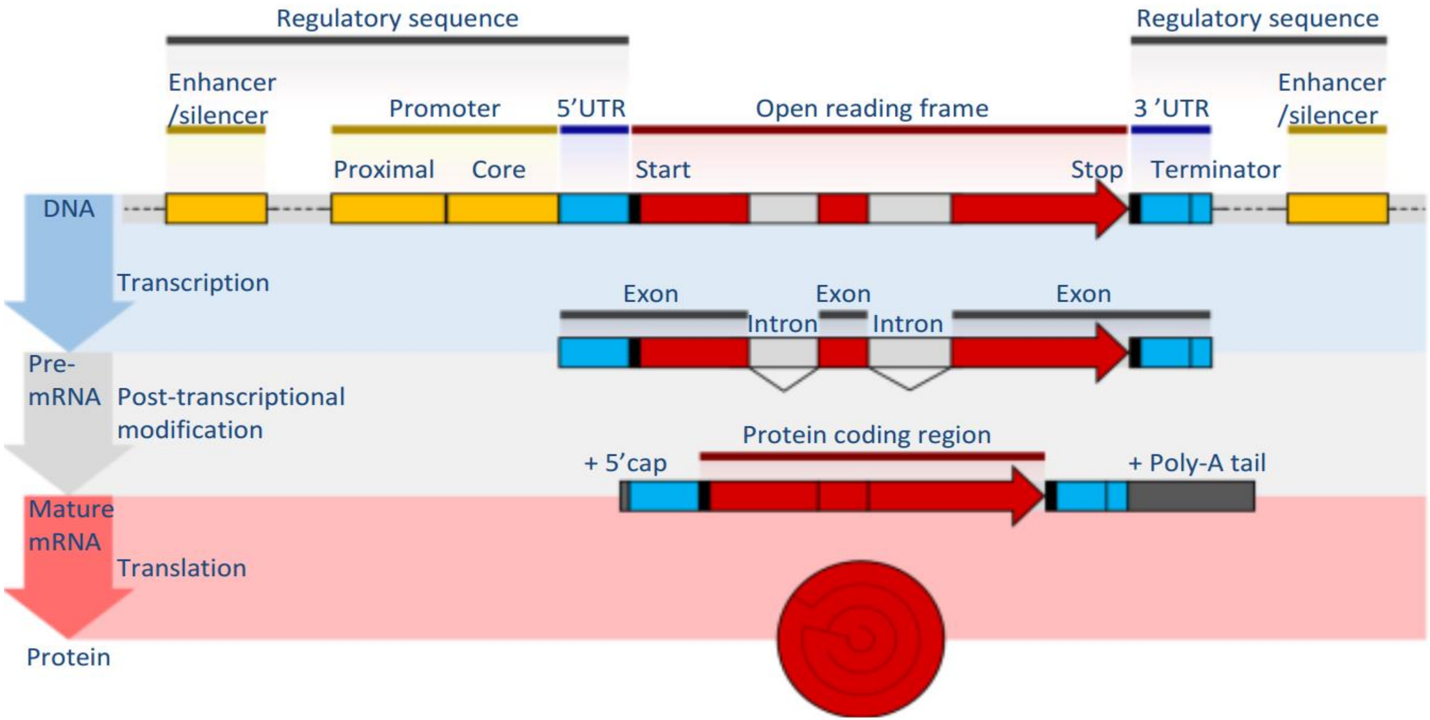
Eukaryotes



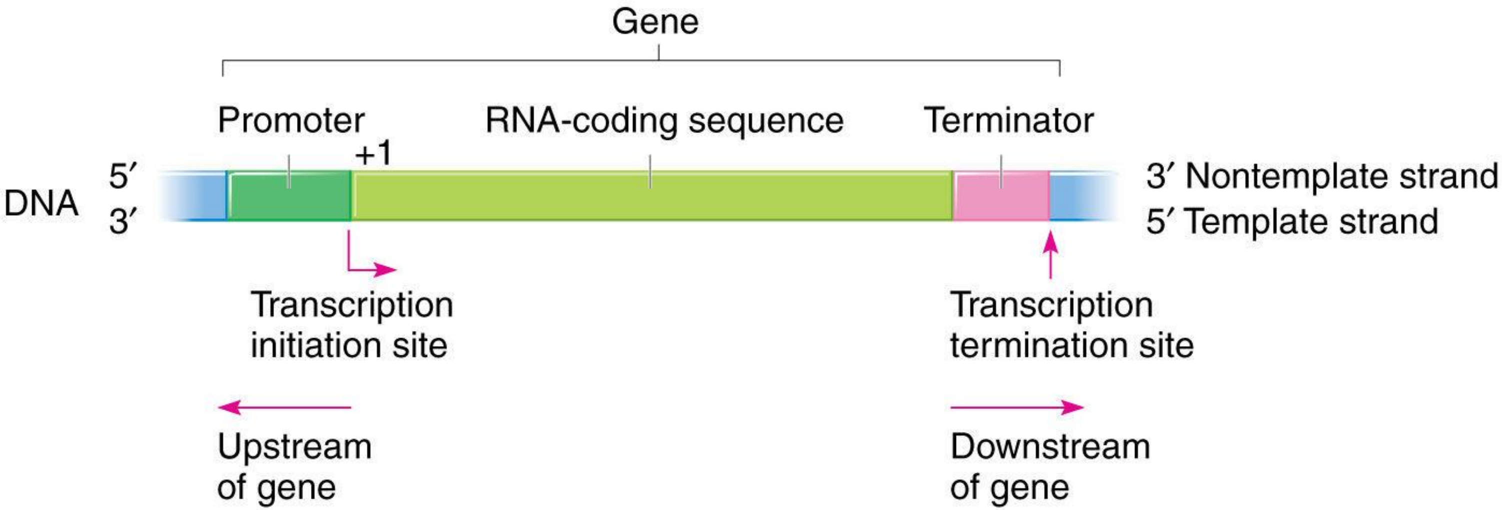
ORF: PROKARYOTES



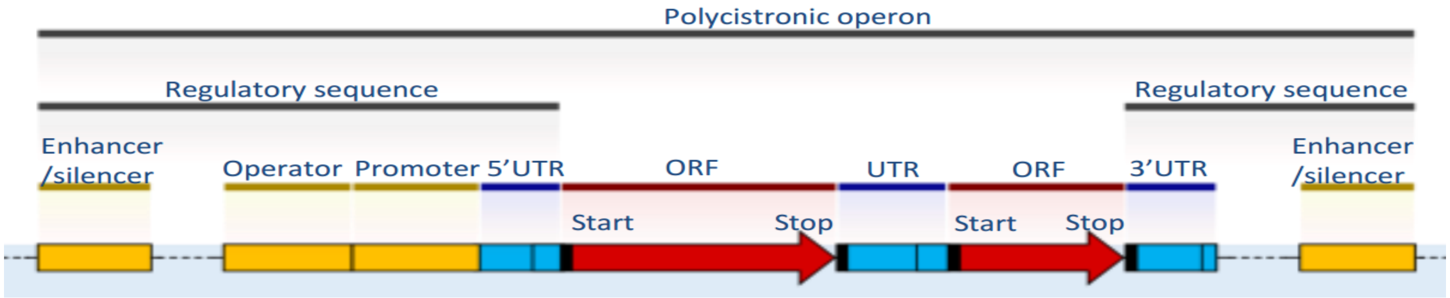
ORF: EUKARYOTES



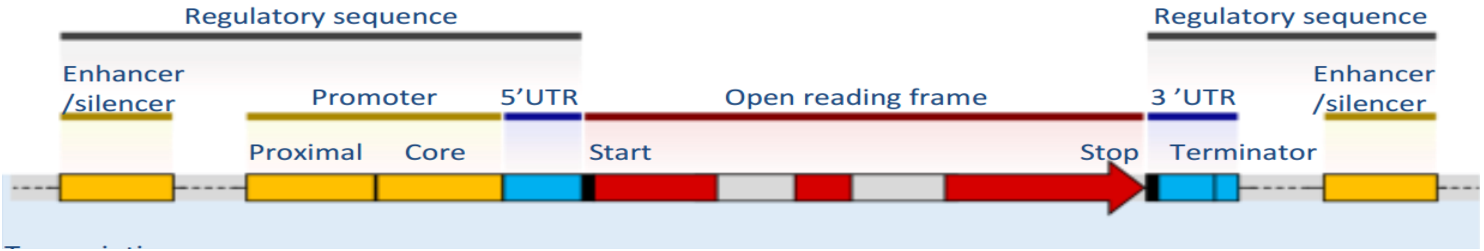
TERMINATOR


Gene
Promoter
RNA-coding sequence
Terminator
+1
DNA
5′
3′
3′ Nontemplate strand
5′ Template strand
Transcription initiation site
Transcription termination site
Upstream of gene
Downstream of gene

Summary: Gene structure

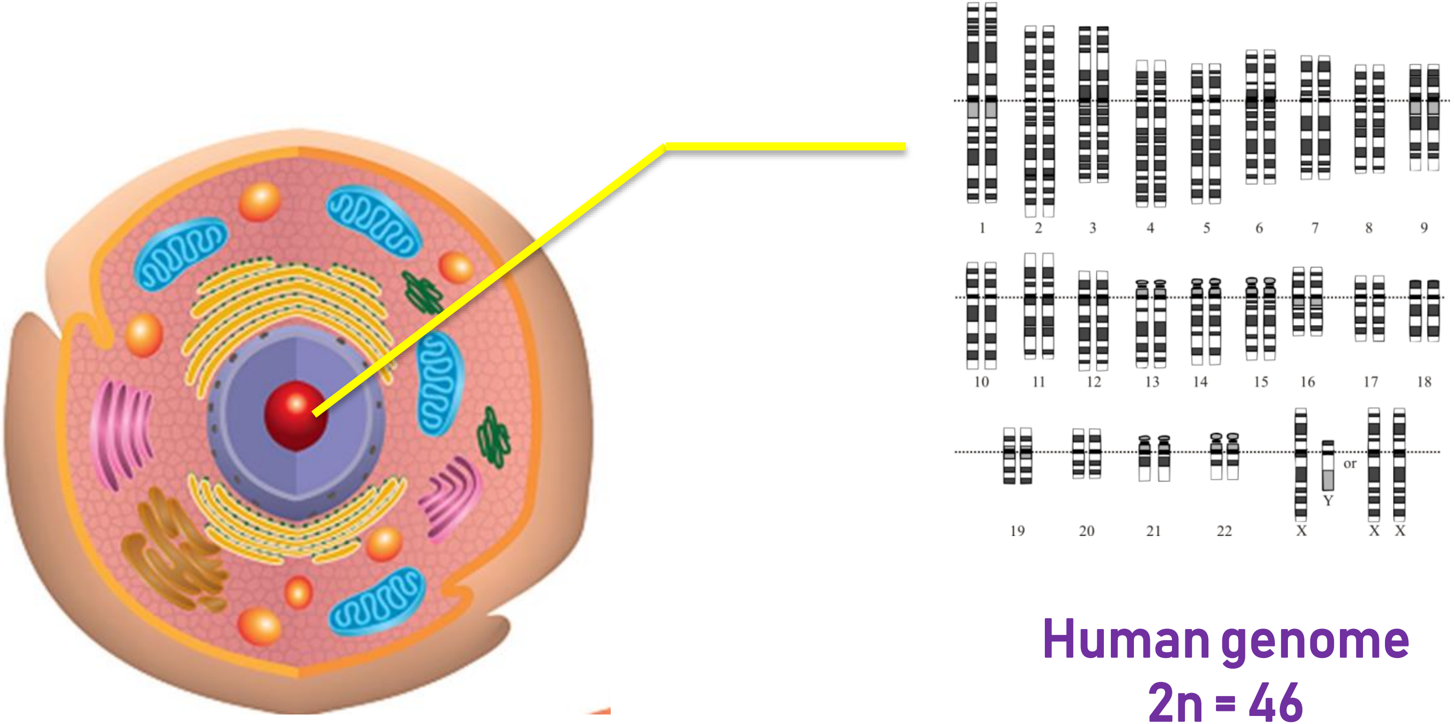


Prokaryotes



Eukaryotes

GENOME

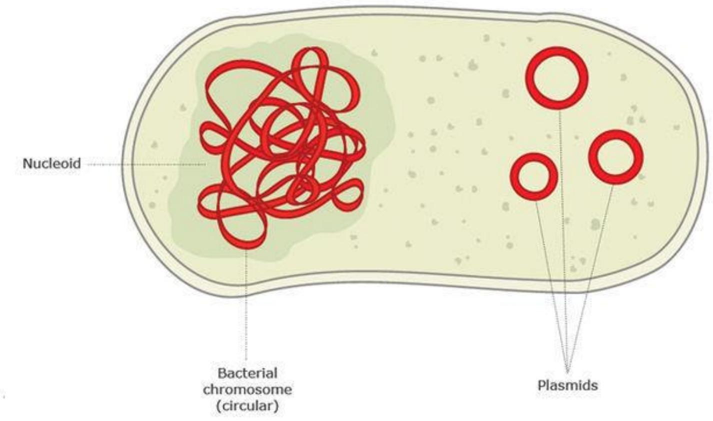


Human genome

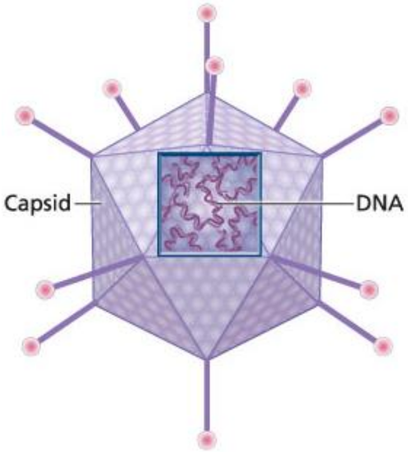
2n = 46

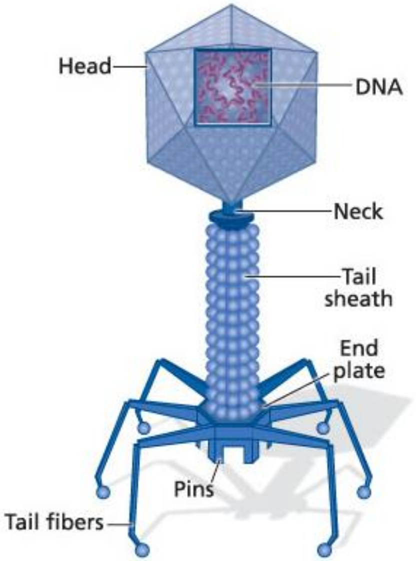
n = 23

GENOME

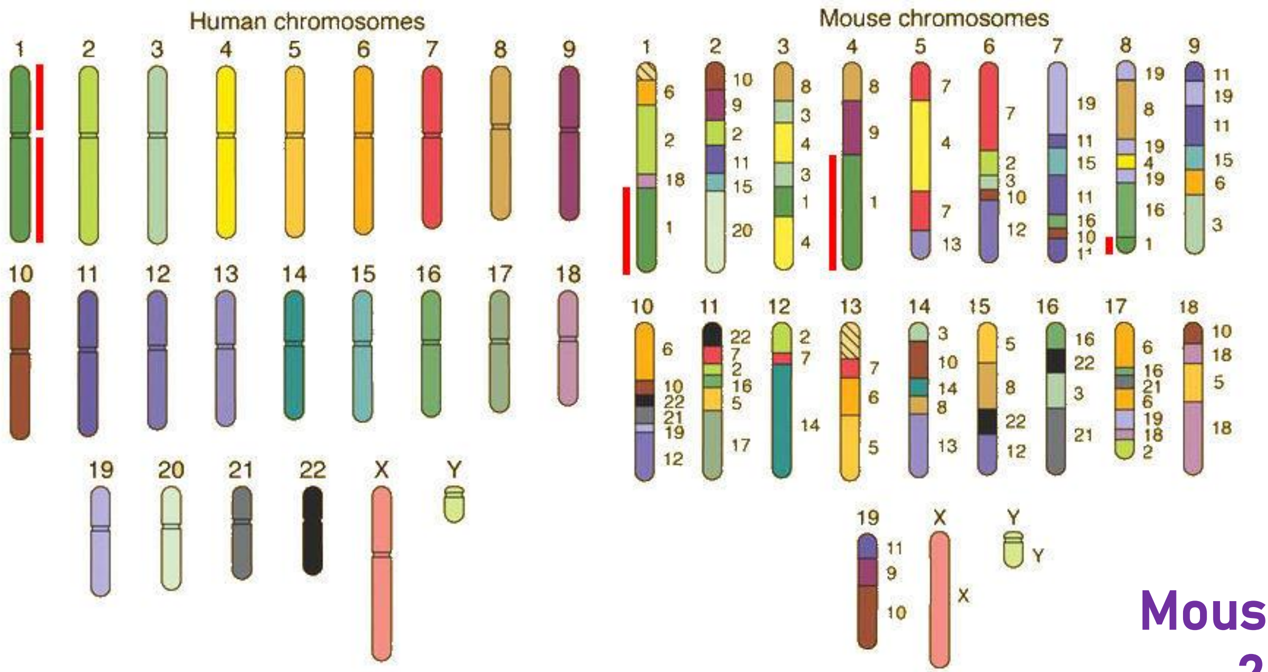


© Copyright. 2014. University of Waikato. All rights reserved.
www.biotechlearn.org.nz



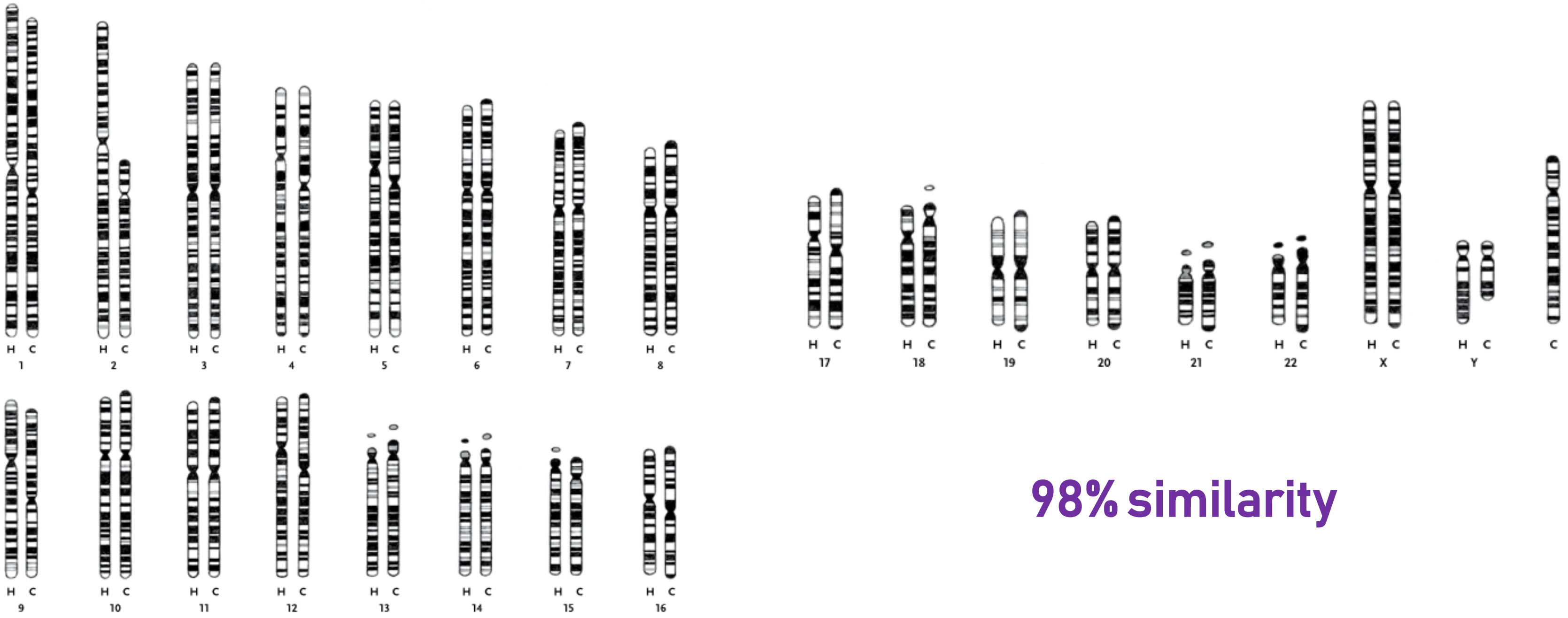


GENOME



Mouse genome
2n = 40

GENOME



98% similarity

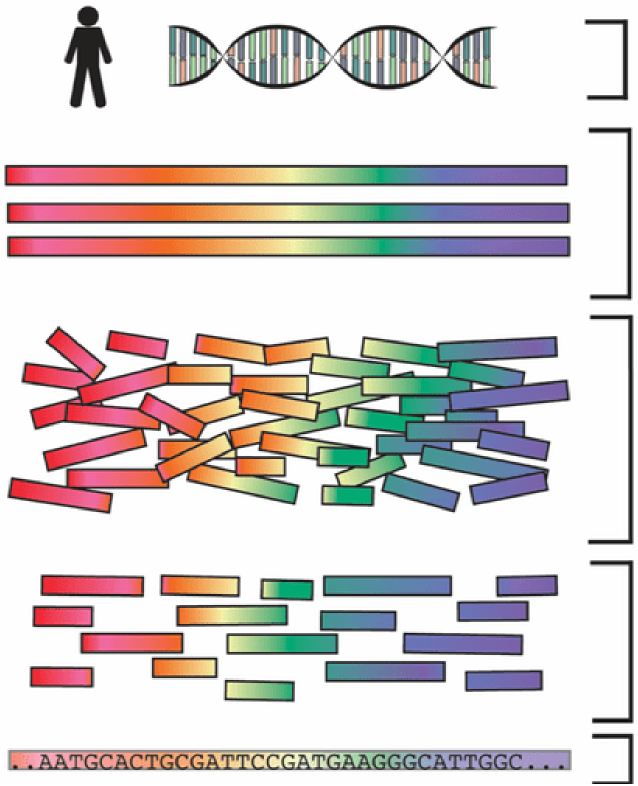
GENOME

So what makes us HUMANS???

→ The need to understand more about human genes

HUMAN GENOME PROJECT

Figure 2: Shotgun Whole-Genome Sequencing



A DNA sample is collected

Many copies of the DNA are made

The copies are broken into many pieces

Sequences are arranged in the correct order

The complete genome is assembled

HUMAN GENOME PROJECT

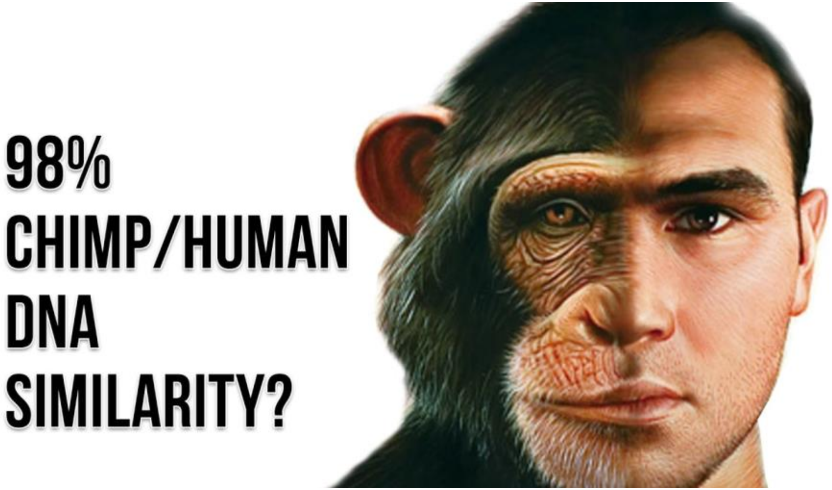
- About 25,000 genes in the human genome
- Regulatory genes
  - promoter sequences
  - regulatory sequences (enhancers, silencers, etc.)
  - intervening sequences (introns)
  - initiation and termination sequences
  - telomeric sequences
  - centromere sequences
  - pseudogenes (have start and stop codons, but are inactive)
  - repetitive DNA
  - transposable genetic elements
- Most mutations occur in males (sperm)
- 3% of genes → code for proteins
- 97% of junk DNA??? **Nothing** to do with genes!!!

→ Under study

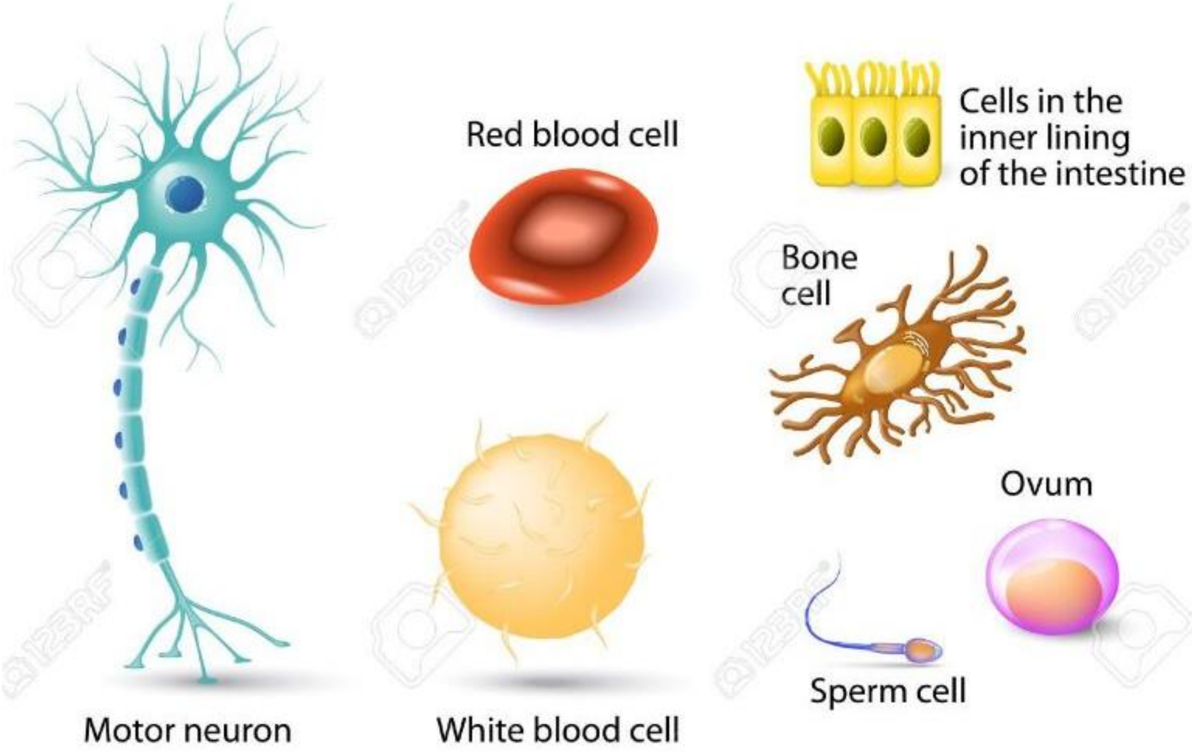
HUMAN GENOME PROJECT

- Compare human genes and those of other organisms → discover gene functions, identify important genes
- Information from DNA → develop new ways to treat, cure, or even prevent diseases
  - Targeted drug/treatment
  - Screening for diseases (Eg: prenatal checkup)

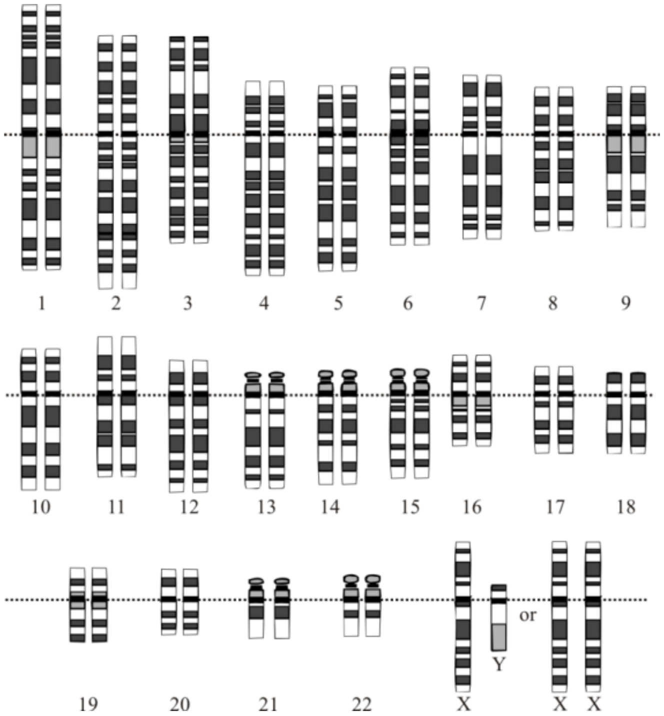

IA 9GAG.COM



99.9% similarity


Red blood cell
Cells in the inner lining of the intestine
Bone cell
Ovum
Sperm cell
Motor neuron
White blood cell

GENETIC VARIATION



- Polymorphism (đa hình)
  - One gene → TWO ALLELES
- Single nucleotide polymorphisms (SNPs) → 1%
- Structural variation (copy-number variation and deletions, inversions, insertions and duplications) → 2,100 to 2,500 structural variations/person
- Gene expression
- Etc.

➔ NO two identical people

➔ Total of 324 million known variants

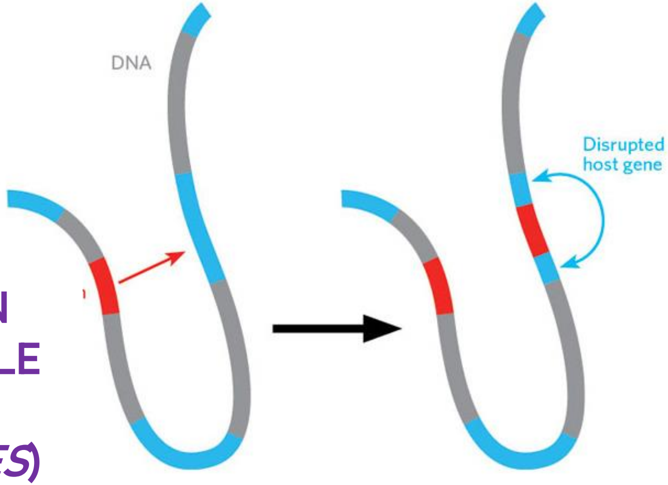
GENETIC VARIATION

- Structural variation
  - Copy-number variation
  - Deletions
  - Inversions
  - Insertions
  - Duplications

GENETIC VARIATION

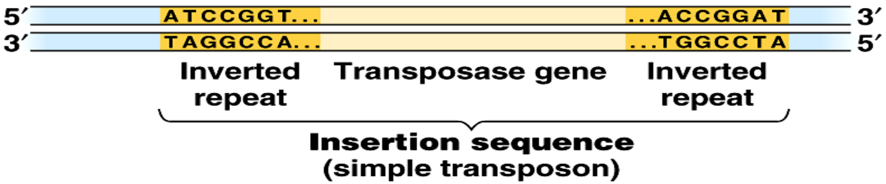
- Structural variation
  - Copy-number variation
  - Deletions
  - Inversions
  - Insertions
  - Duplications

TRANSPOSON (TRANSPOSABLE ELEMENT = *JUMPING GENES*)

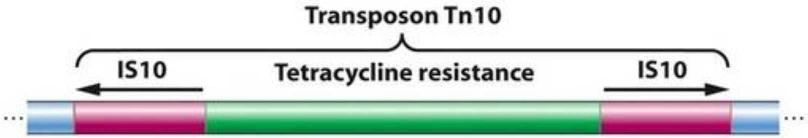


TRANSPOSABLE ELEMENTS (TEs)

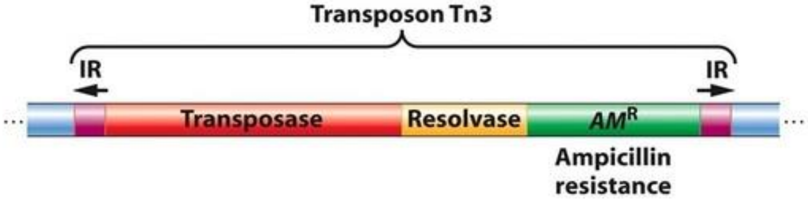
INSERTION SEQUENCE (IS)



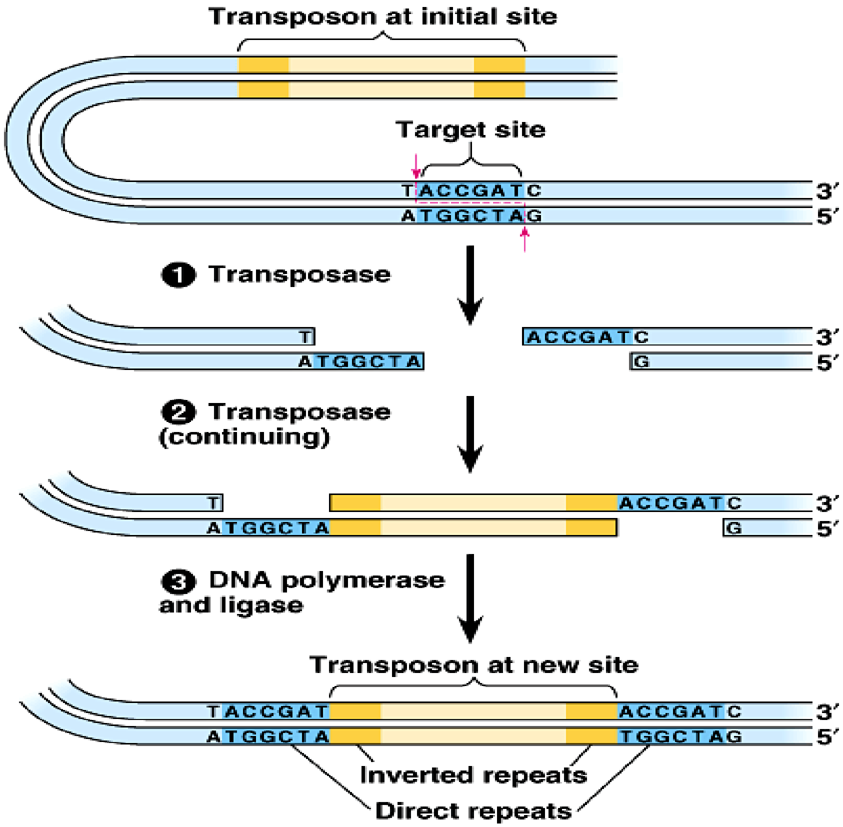
(a) Composite transposon



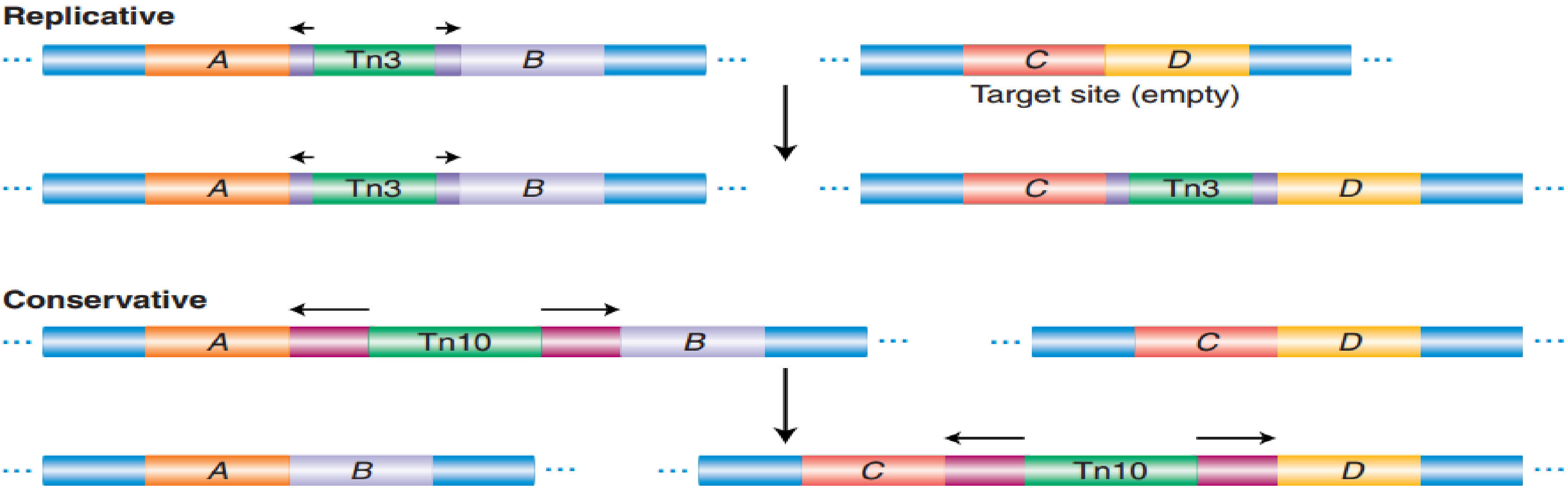
(b) Simple transposon (noncomposite)



MECHANISM


Transposon at initial site
Target site
TACCGATC
ATGGCTAG
3′
5′
1 Transposase
T
ATGGCTA
ACCGATC
G
3′
5′
2 Transposase (continuing)
T
ATGGCTA
ACCGATC
G
3′
5′
3 DNA polymerase and ligase
Transposon at new site
TACCGAT
ATGGCTA
ACCGATC
TGGCTAG
3′
5′
Inverted repeats
Direct repeats

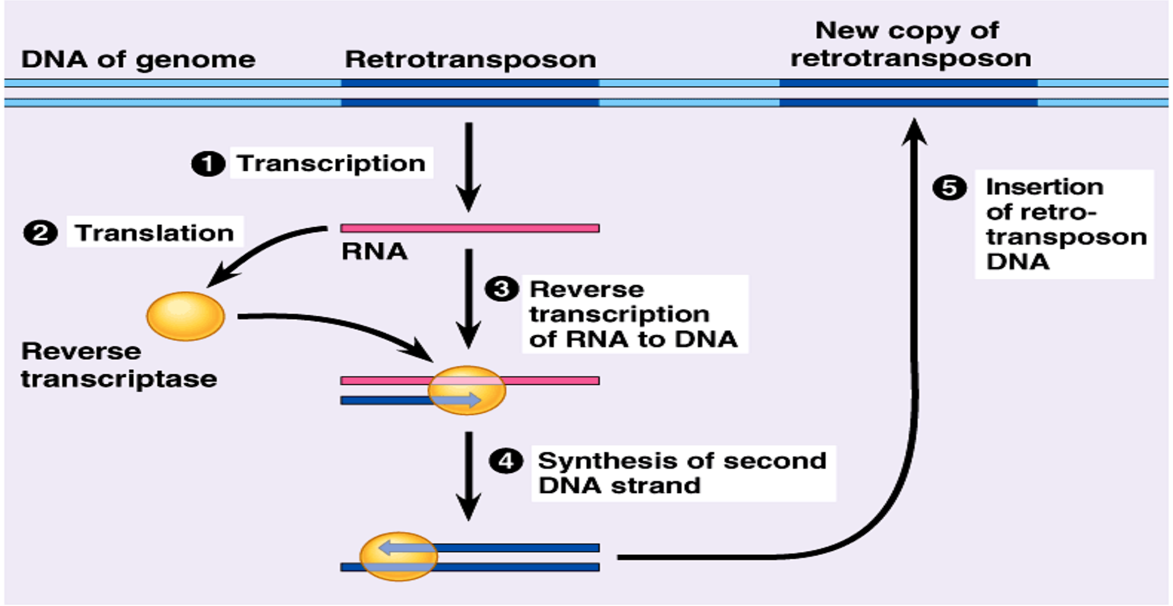
TWO BASIC METHODS



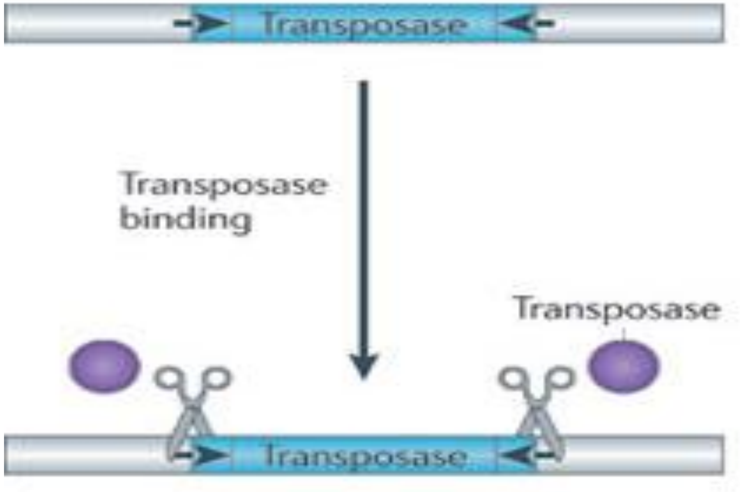
TRANSPOSABLE ELEMENTS (TEs)

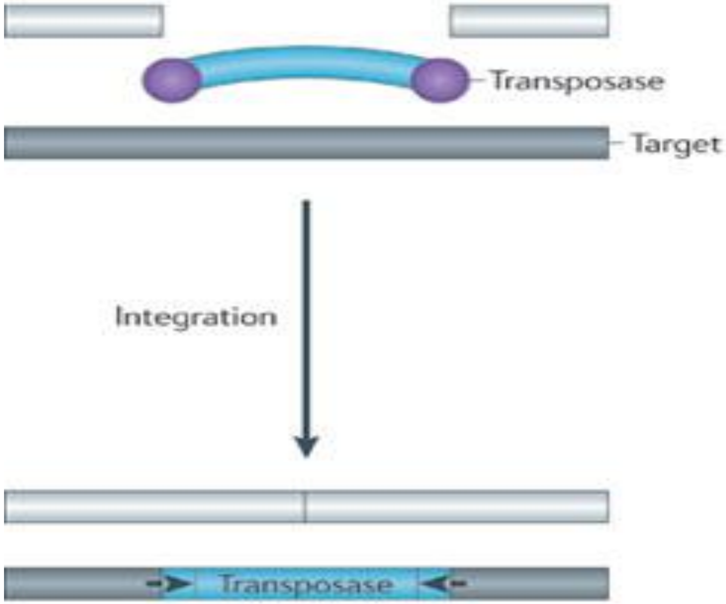
- TEs come in many different forms and shapes
  - Retrotransposons (RNA intermediate)
  - DNA transposons (DNA intermediate)
- Balance between TE expression and repression

CLASS I: RETROTRANSPOSON
RNA INTERMEDIATE



CLASS II: DNA TRANSPOSON
DNA INTERMEDIATE



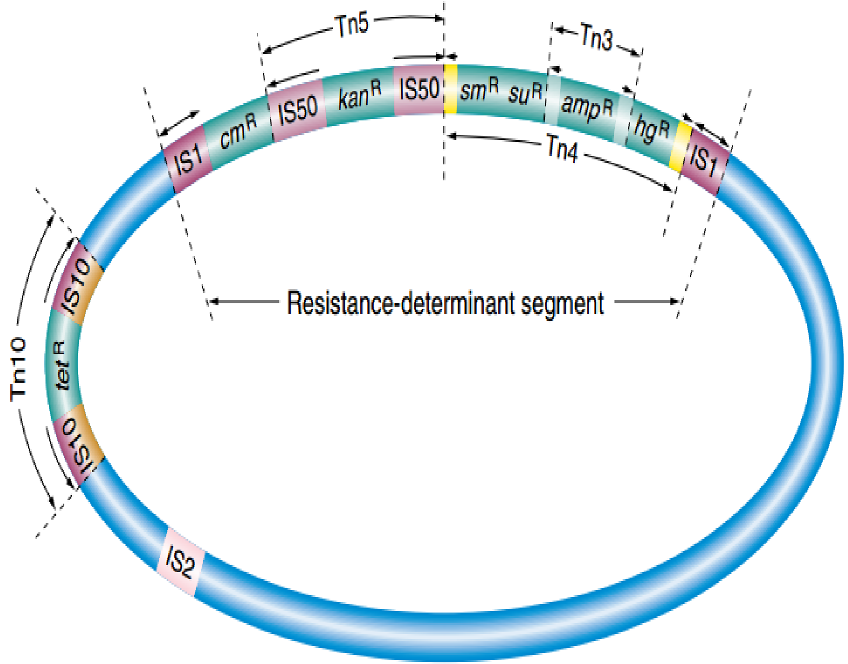


TRANSPOSONS: PROKARYOTE

- Plasmid → bacteria
- Transposon plasmid

→ bacterial chromosome /plasmid

- Multiple drug-resistant plasmids

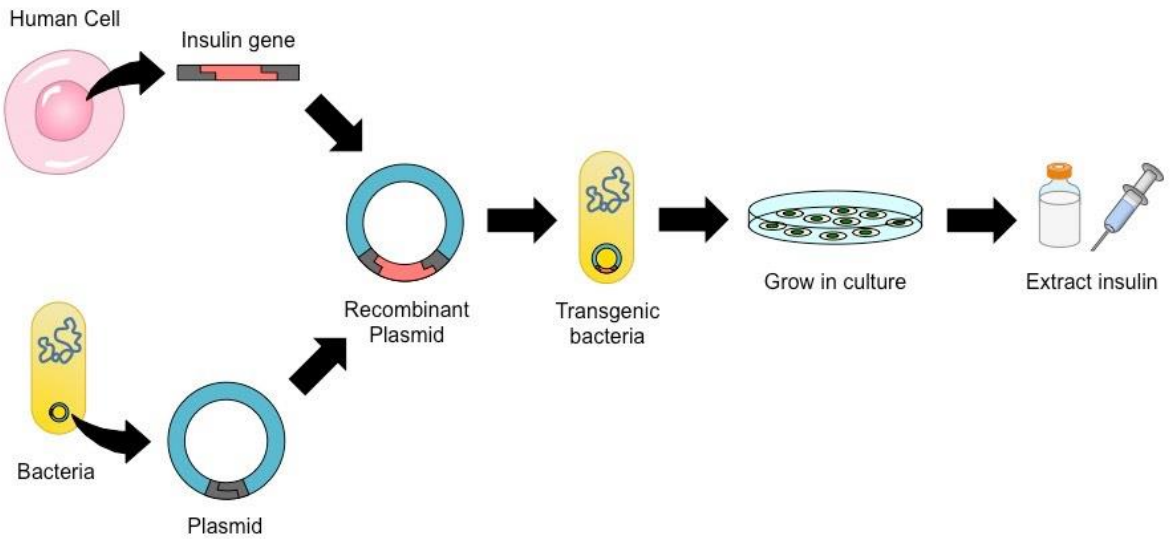


TRANSPOSONS: EUKARYOTE

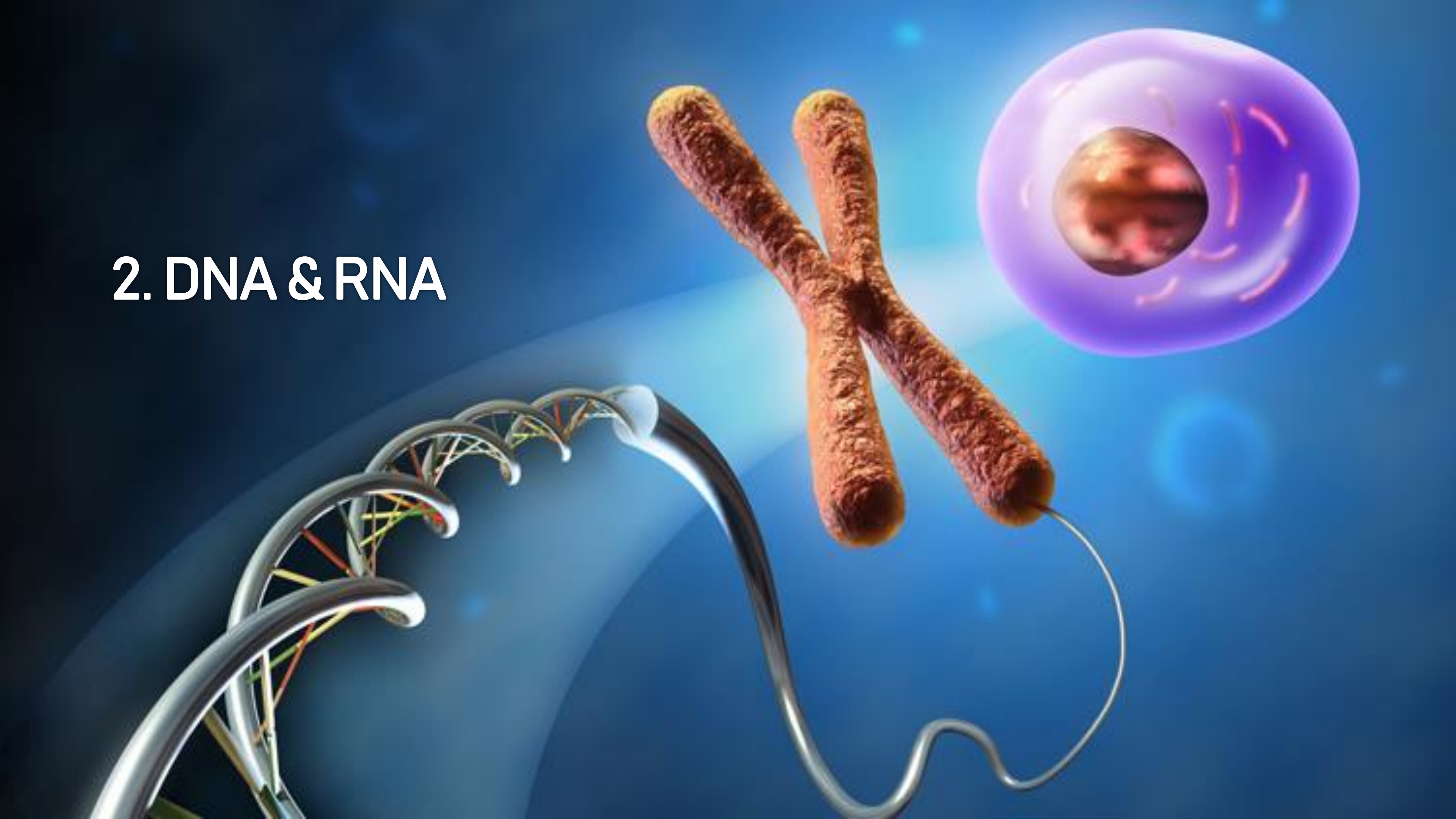
- TEs are not randomly distributed in the genome
- TEs are an extensive source of mutations and genetic polymorphisms
- TEs are associated with genome rearrangements and unique chromosome features

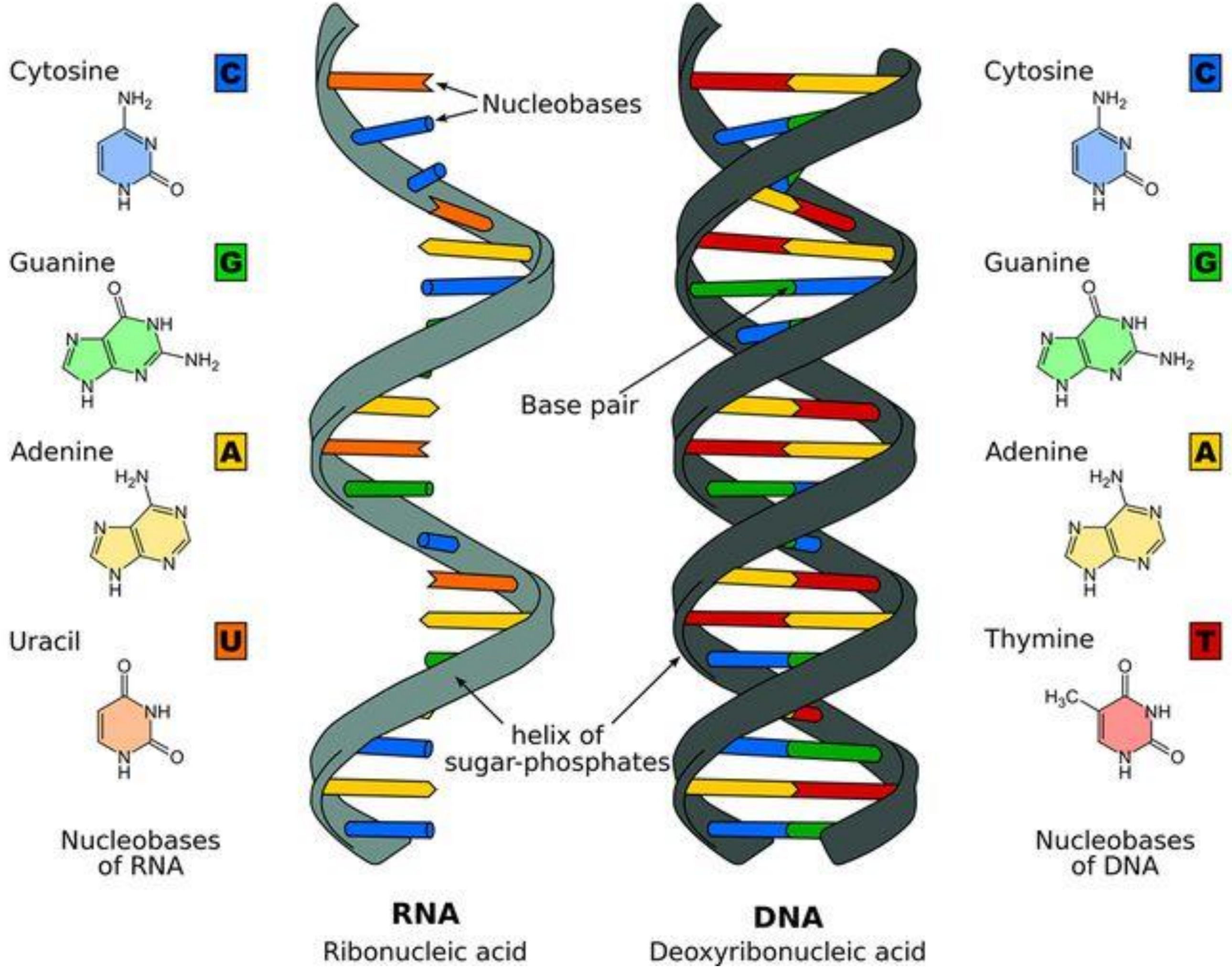
TRANSPOSONS APPLICATION

- Induce mutations (Insertional mutagenesis)
- Genetic modification
- Transformation vectors for transferring genes between organisms, developing plasmid to transfer genes



2. DNA & RNA



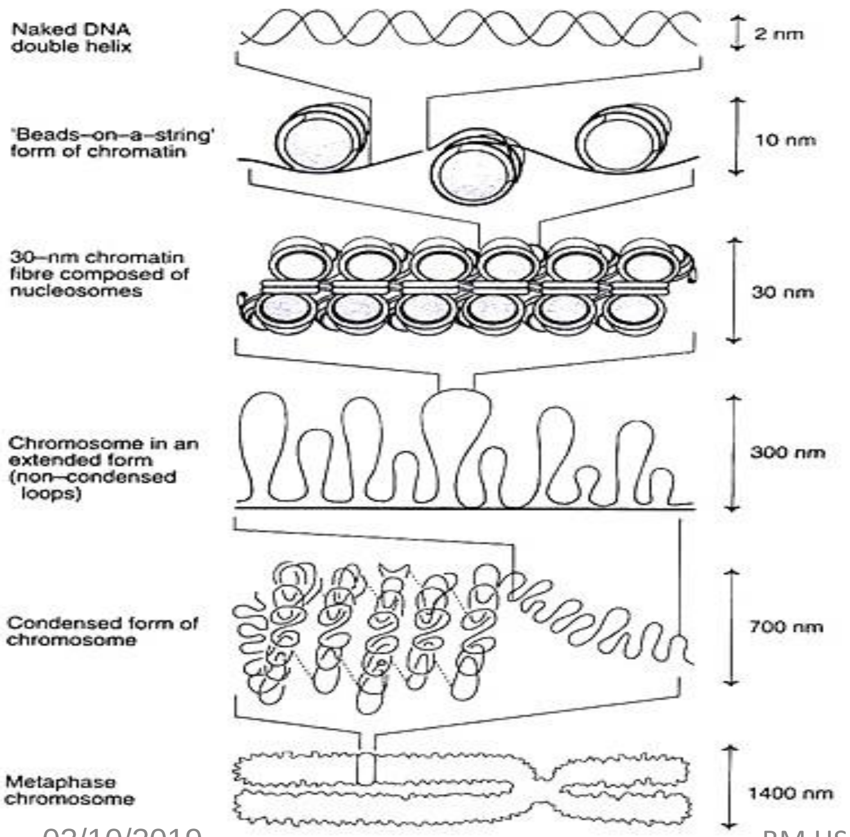

Cytosine
C
Guanine
G
Adenine
A
Uracil
U
Nucleobases of RNA
Nucleobases
Base pair
helix of sugar-phosphates
RNA
Ribonucleic acid
DNA
Deoxyribonucleic acid
Cytosine
C
Guanine
G
Adenine
A
Thymine
T
Nucleobases of DNA

DNA & RNA SIMILARITIES

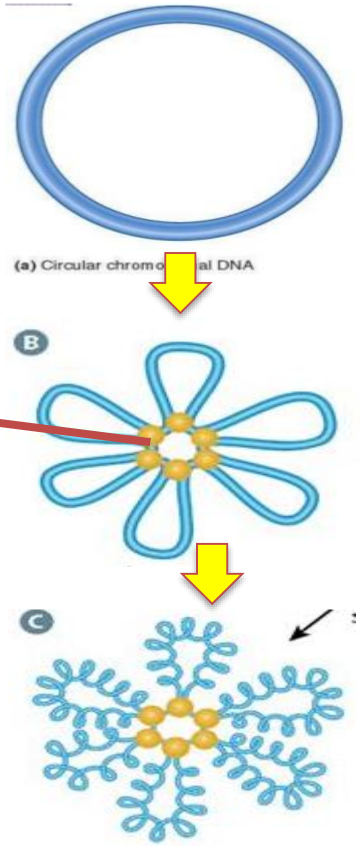
- Polymers of nucleotides. Nucleic acids. Phosphate backbone
- Genetic materials, passing inheritance information

	DNA	RNA
Base	Thymine	Uracil
Strand	Double	Single
Sugar	Deoxyribose (Less reactive)	Oxyribose (More reactive)
Length	Long	Short
Lifespan	Longer	Shorter
Function	Store genetic information	Passing information
Location (Eukaryote)	Nucleus	Cytoplasm

Eukaryotes



Prokaryotes



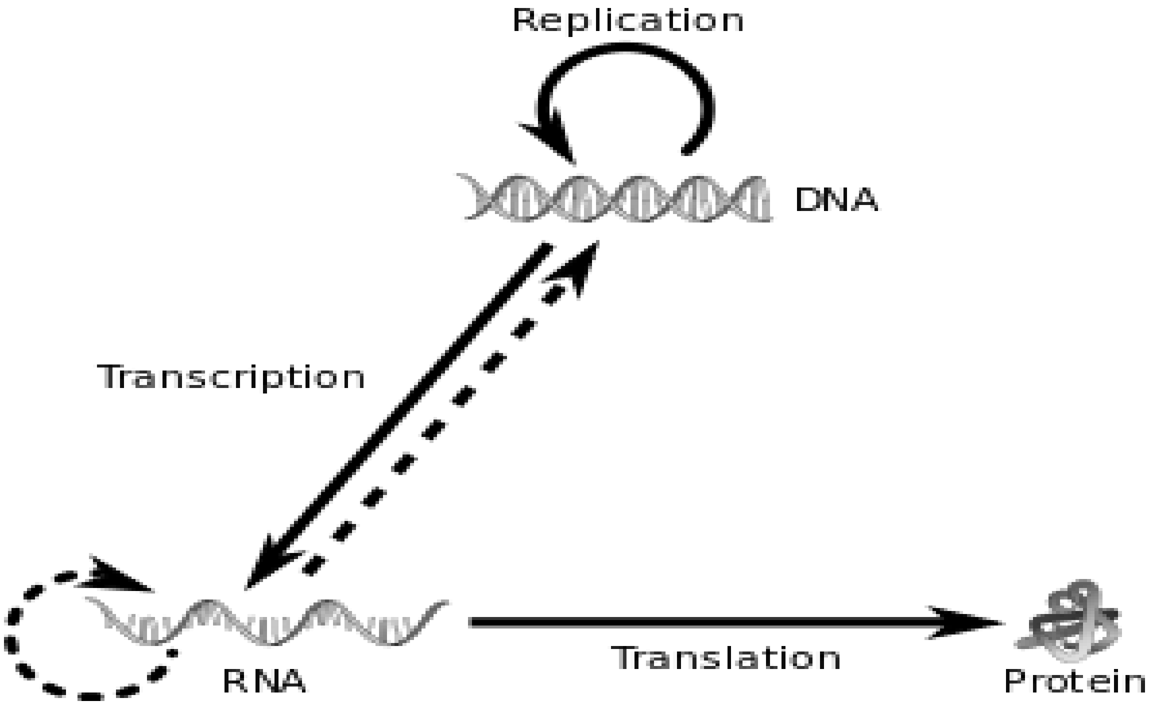
Circular
Unfolded
chromosome

protein

Folded,
looped
chromosome

Folded,
supercoiled
chromosome

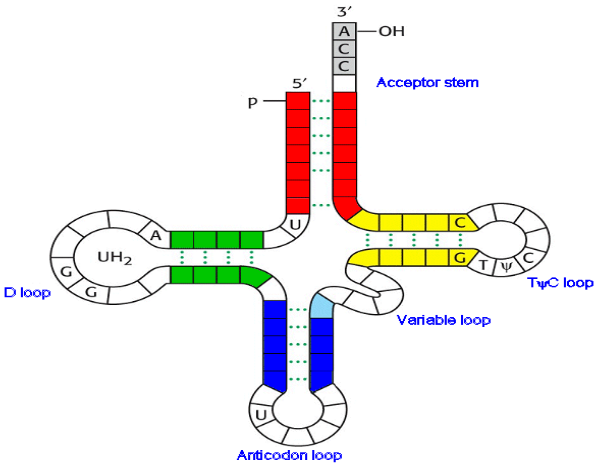
Central Dogma



DIFFERENT TYPES OF RNAS

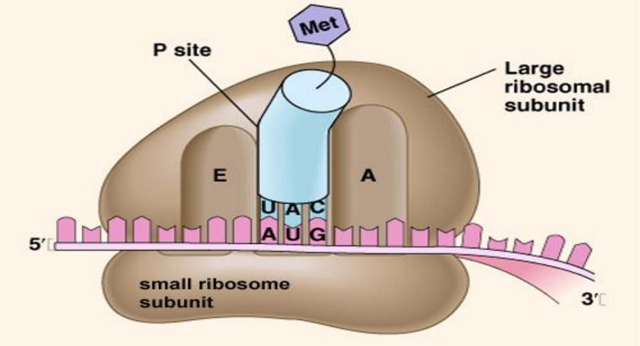
- mRNA: messenger RNA
  - 2%
  - mRNA transcribes the genetic code from DNA into a form that can be read and used to make proteins
  - Eukaryotes: mRNA carries genetic information from the nucleus to the cytoplasm of a cell → premature RNA → mRNA

DIFFERENT TYPES OF RNAS



- tRNA: transfer RNA
  - Cellular cytoplasm
  - Involved in protein synthesis
  - Transfer RNA brings amino acids to the ribosome that corresponds to each three-nucleotide codon of rRNA. The amino acids then can be joined together and processed to make polypeptides and proteins.

DIFFERENT TYPES OF RNAS

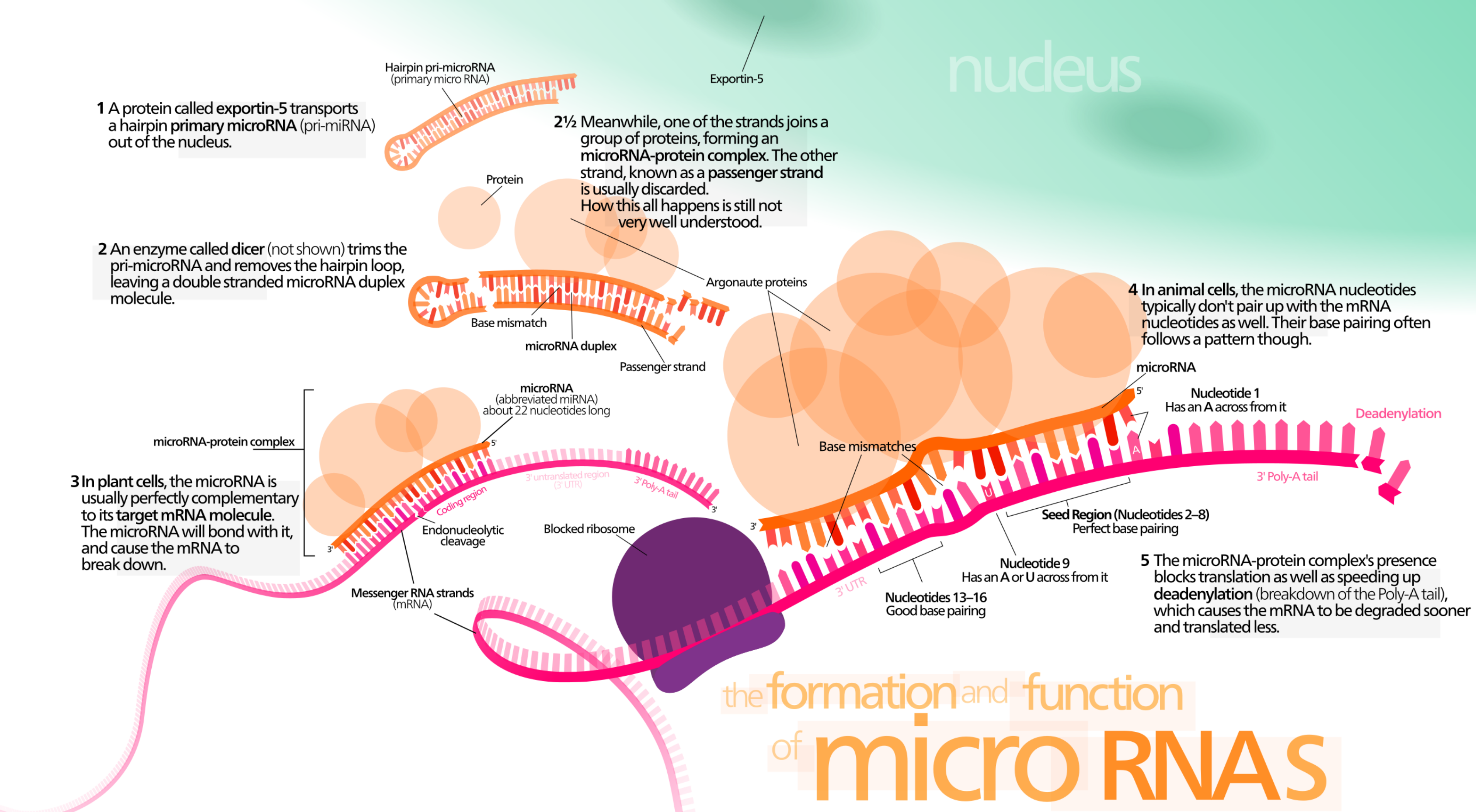


- rRNA: ribosomal RNA
  - Cellular cytoplasm
  - rRNA directs the translation of mRNA into proteins

DIFFERENT TYPES OF RNAS

- miRNA: micro RNA
  - Short non-coding RNAs that regulate gene expression post-transcriptionally
  - Bind to the 3'-UTR (untranslated region) of their target mRNAs and repress protein production by destabilizing the mRNA and translational silencing.

the formation and function of micro RNAs



1 A protein called **exportin-5** transports a hairpin **primary microRNA** (pri-miRNA) out of the nucleus.

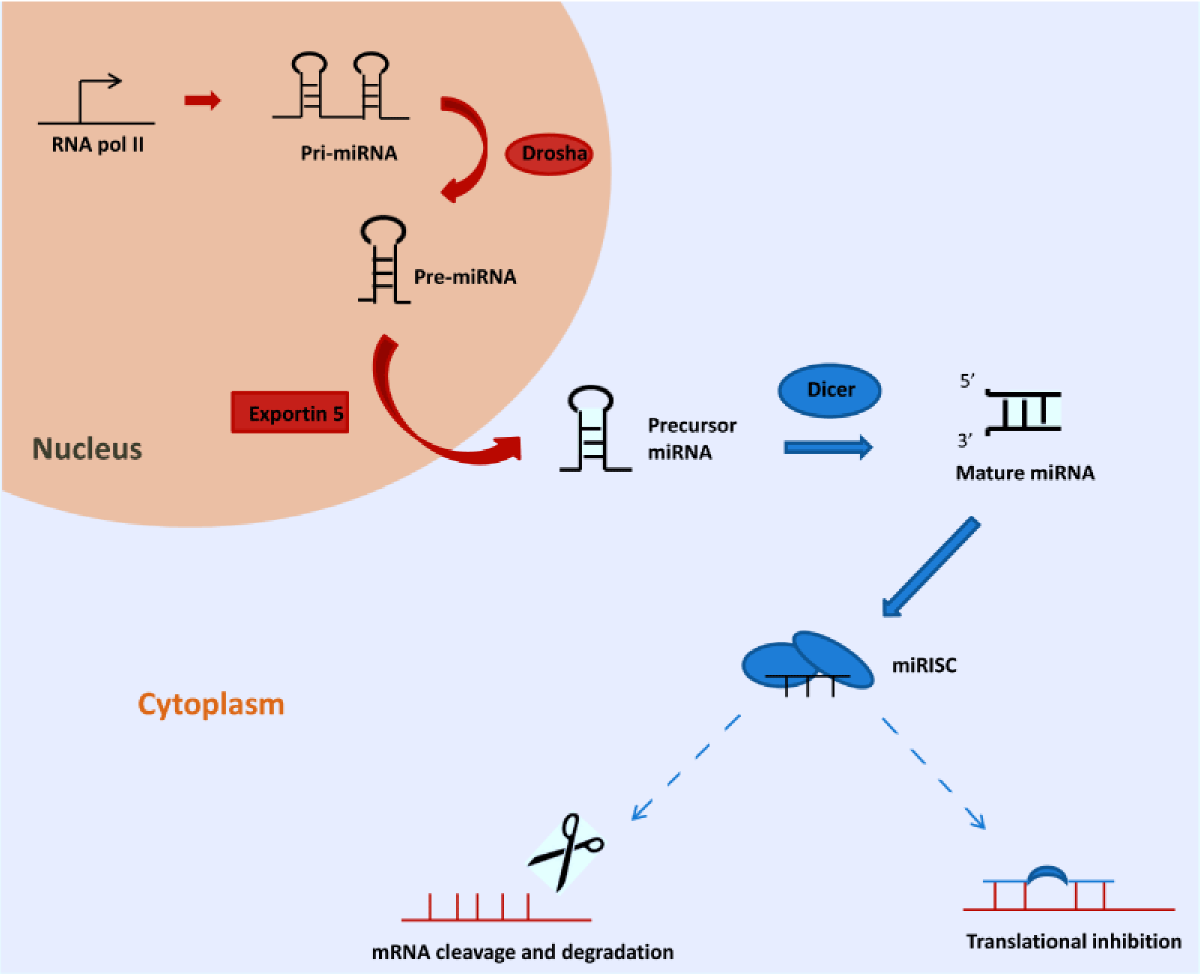
2 An enzyme called **dicer** (not shown) **trims the** pri-microRNA and removes the hairpin loop, leaving a double stranded microRNA duplex molecule.

2½ Meanwhile, one of the strands joins a group of proteins, forming an **microRNA-protein complex.** The other strand, known as a **passenger strand** is usually discarded.
How this all happens is still not very well understood.

3 In plant cells, the microRNA is usually perfectly complementary to its **target mRNA molecule.** The microRNA will bond with it, and cause the mRNA to break down.

4 In animal cells, the microRNA nucleotides typically don't pair up with the mRNA nucleotides as well. Their base pairing often follows a pattern though.

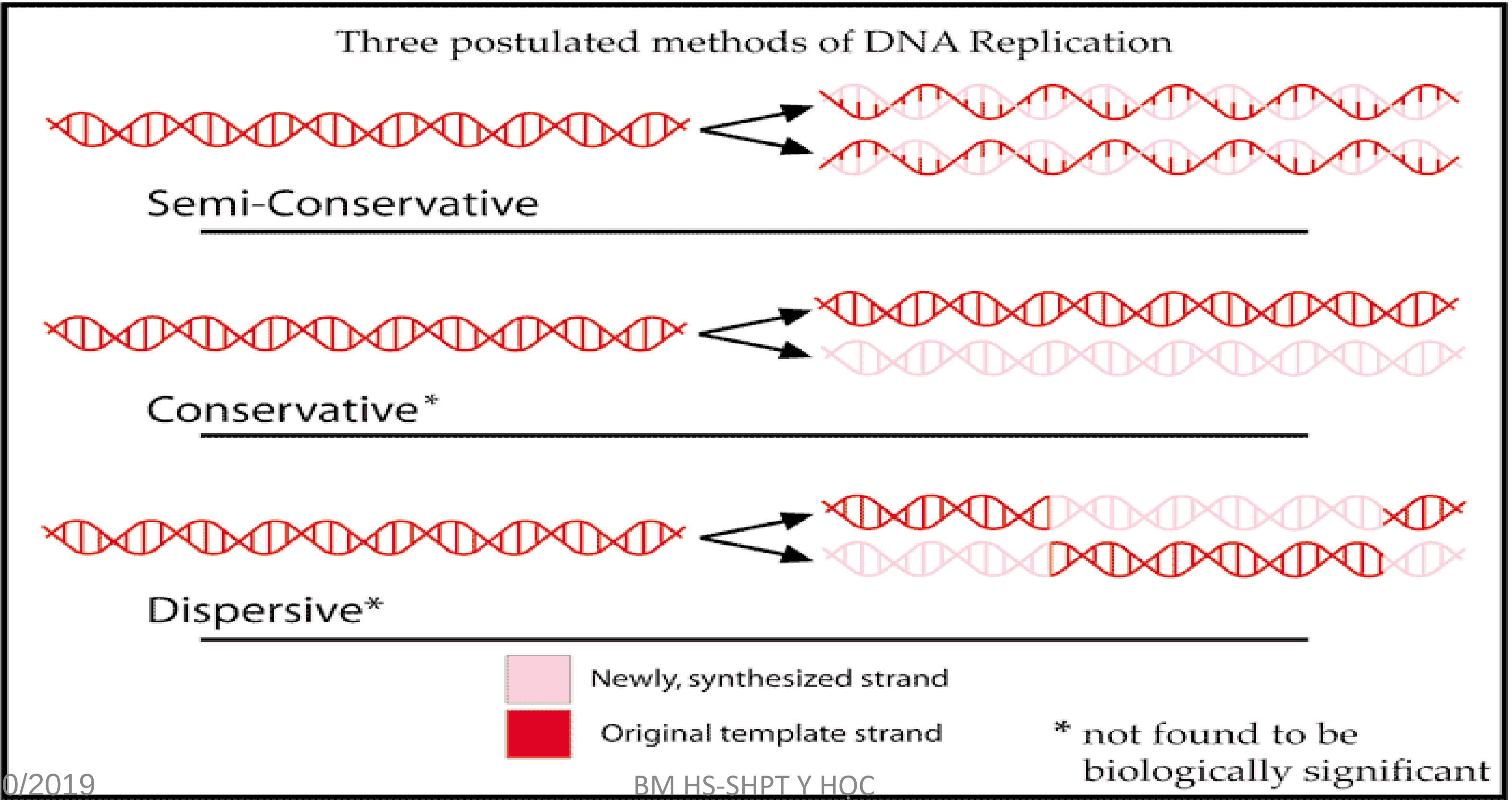
5 The microRNA-protein complex's presence blocks translation as well as speeding up **deadenylation** (breakdown of the Poly-A tail), which causes the mRNA to be degraded sooner and translated less.


RNA pol II
Pri-miRNA
Drosha
Pre-miRNA
Exportin 5
Nucleus
Precursor miRNA
Dicer
5′
3′
Mature miRNA
miRISC
Cytoplasm
mRNA cleavage and degradation
Translational inhibition

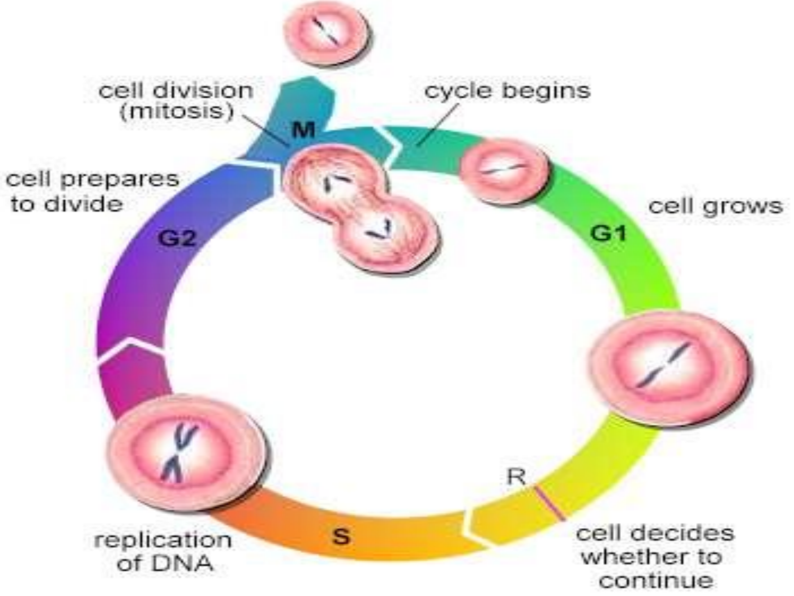
3. GENETIC PROCESSES

DNA REPLICATION

SEMICONSERVATIVE



DNA REPLICATION



i. Quá trình bán bảo tồn (Semi-conservative)

ii. Gốc sao chép (origins of replication - ori)

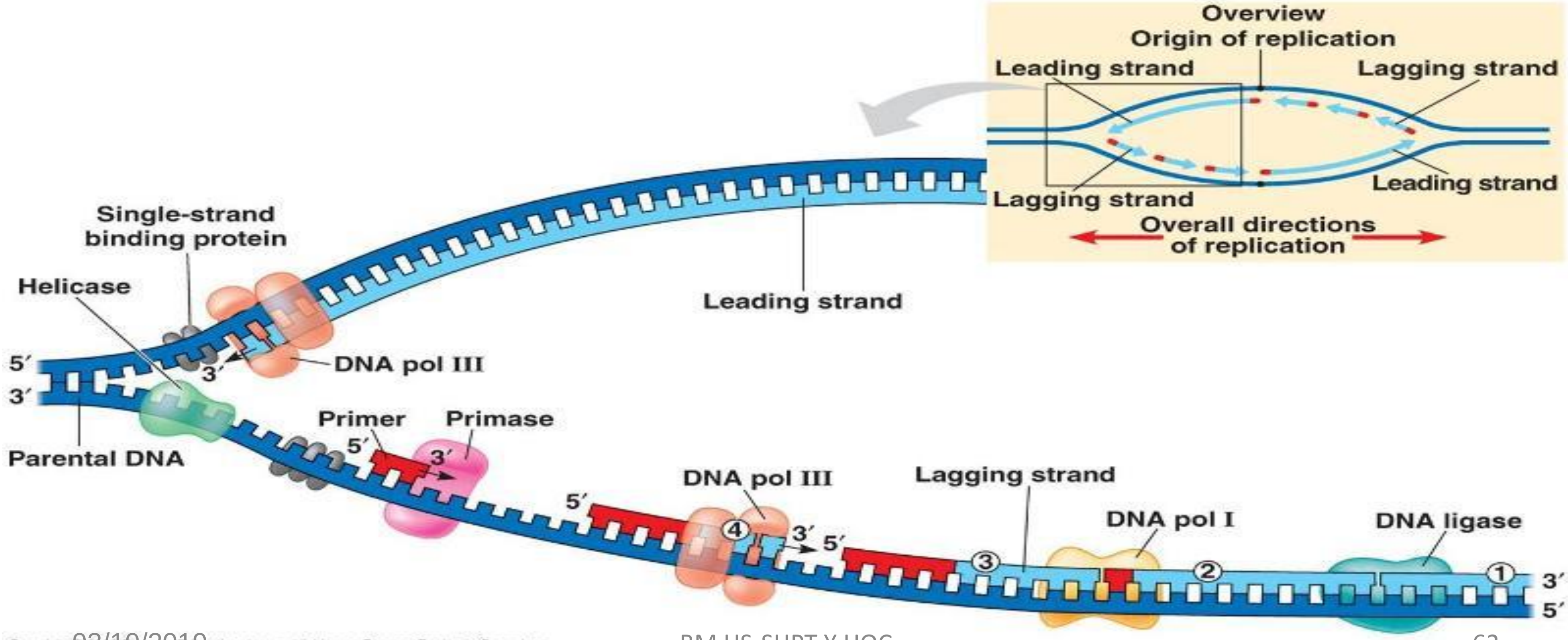
iii. Chiều 5' → 3'

iv. Gián đoạn ở một trong hai chuỗi

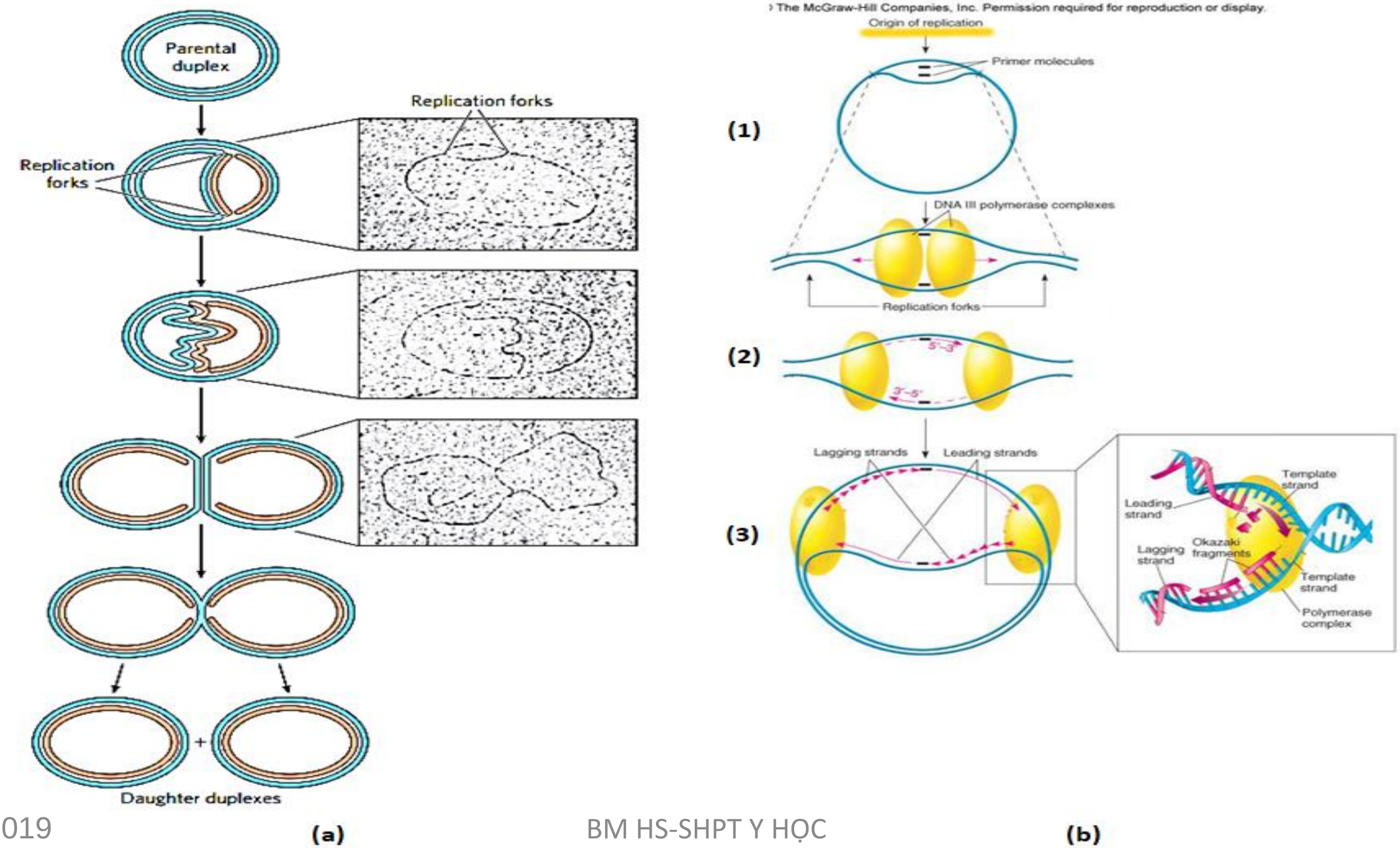
v. “Mồi“ (RNA primer)

vi. Protein enzyme đặc hiệu.

DNA REPLICATION – Prokaryote

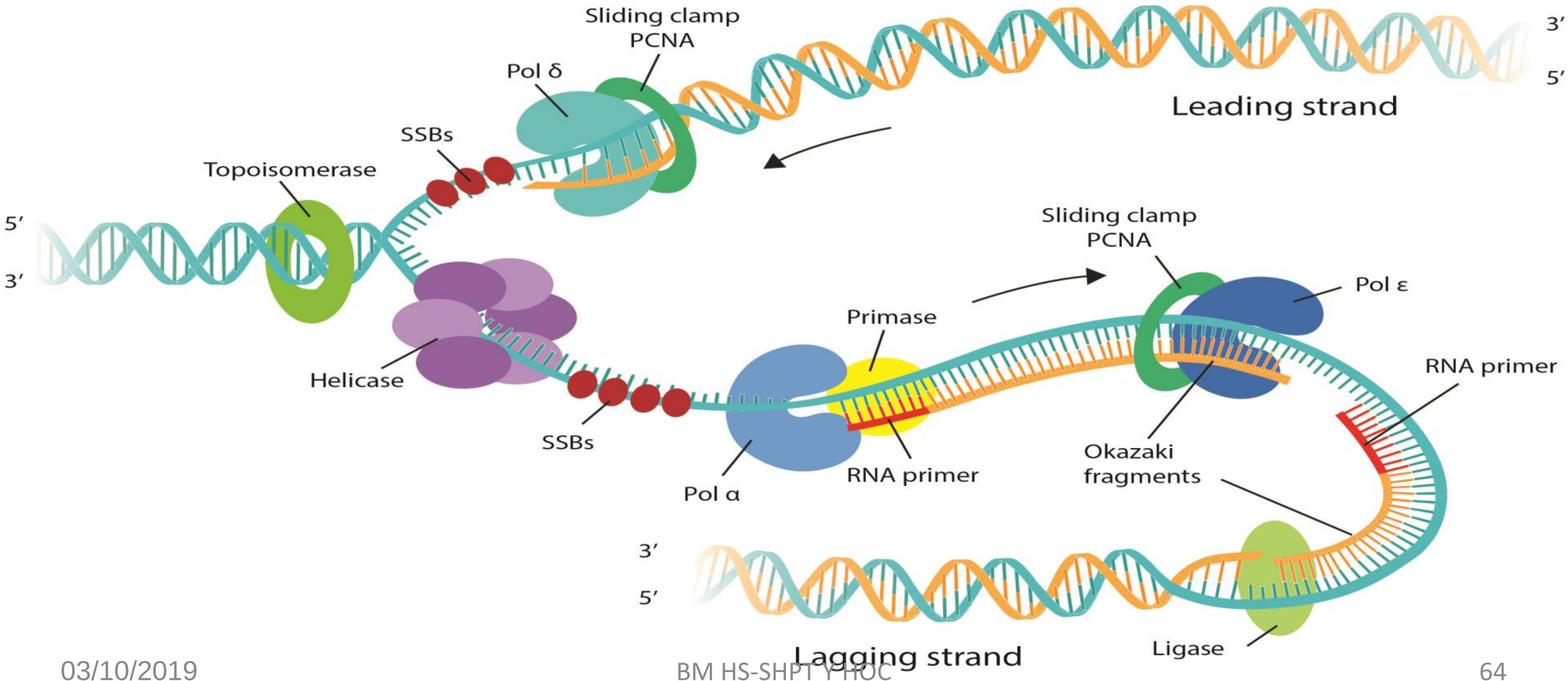


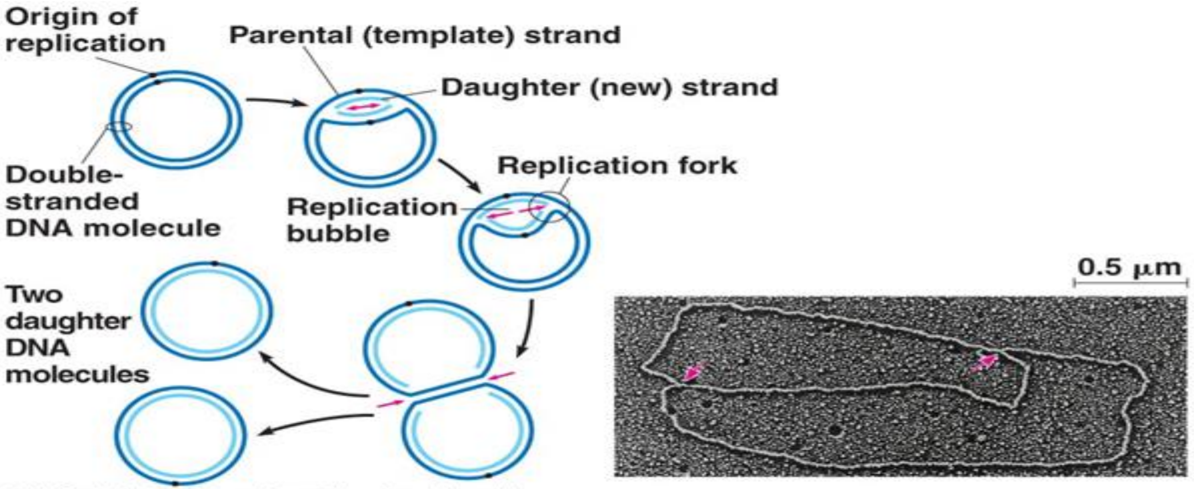
DNA REPLICATION – Prokaryote



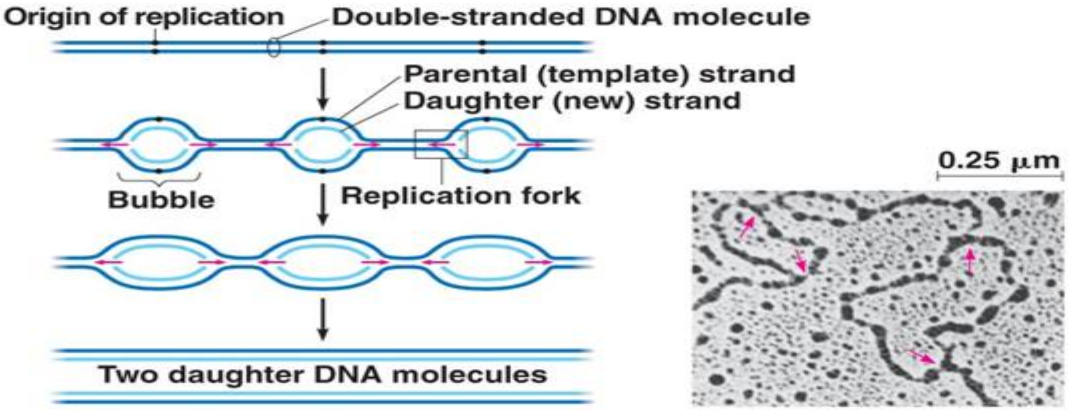
) The McGraw-Hill Companies, Inc. Permission required for reproduction or display.

DNA REPLICATION – Eukaryote





(a) Origins of replication in *E. coli*

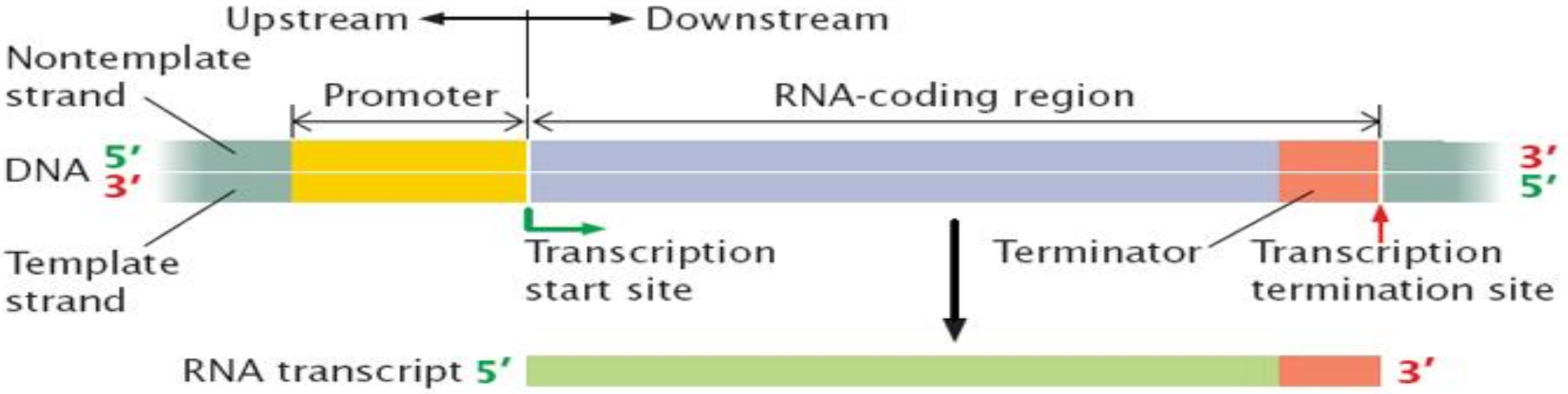


(b) Origins of replication in eukaryotes

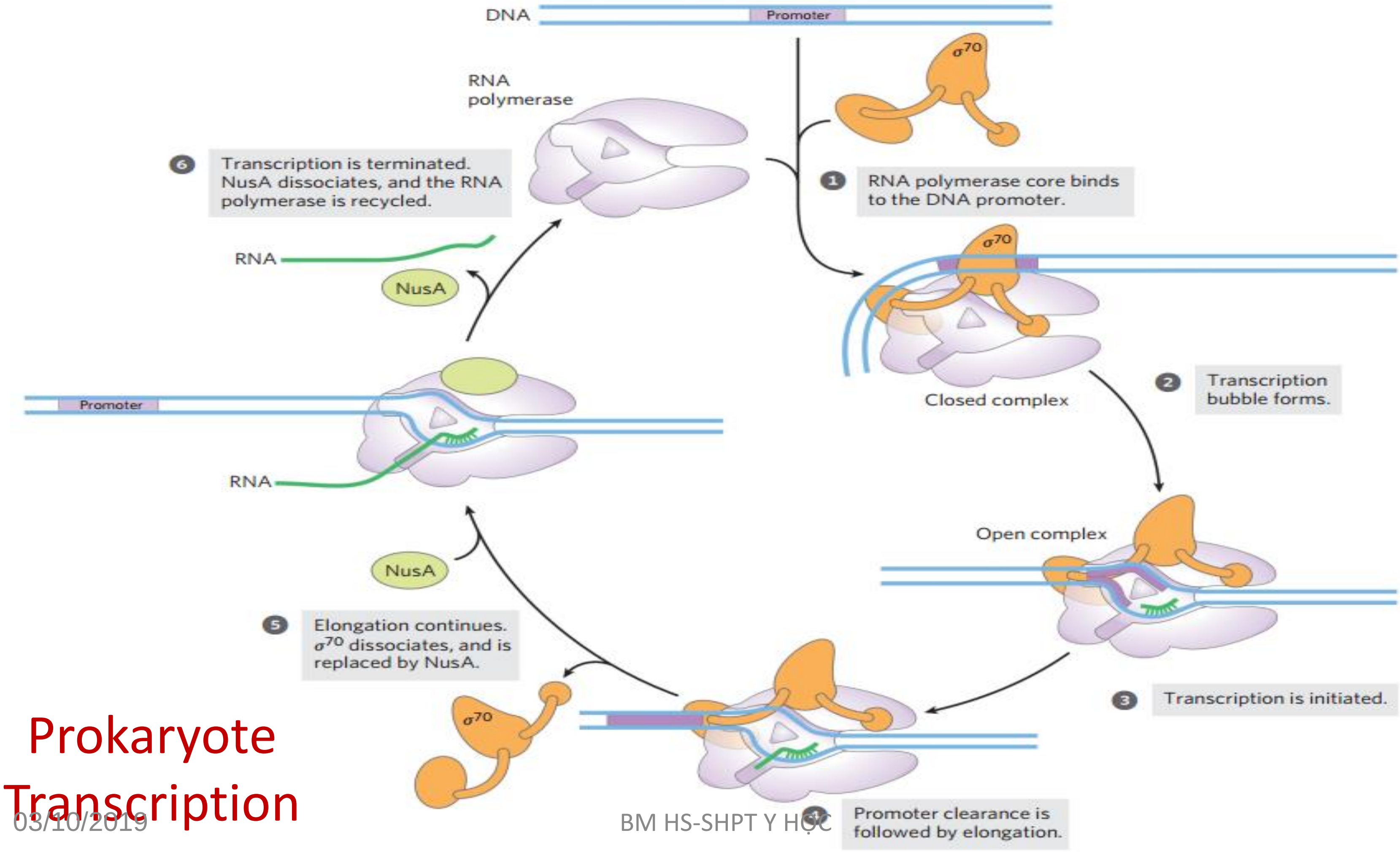
TRANSCRIPTION

Đơn vị phiên mã

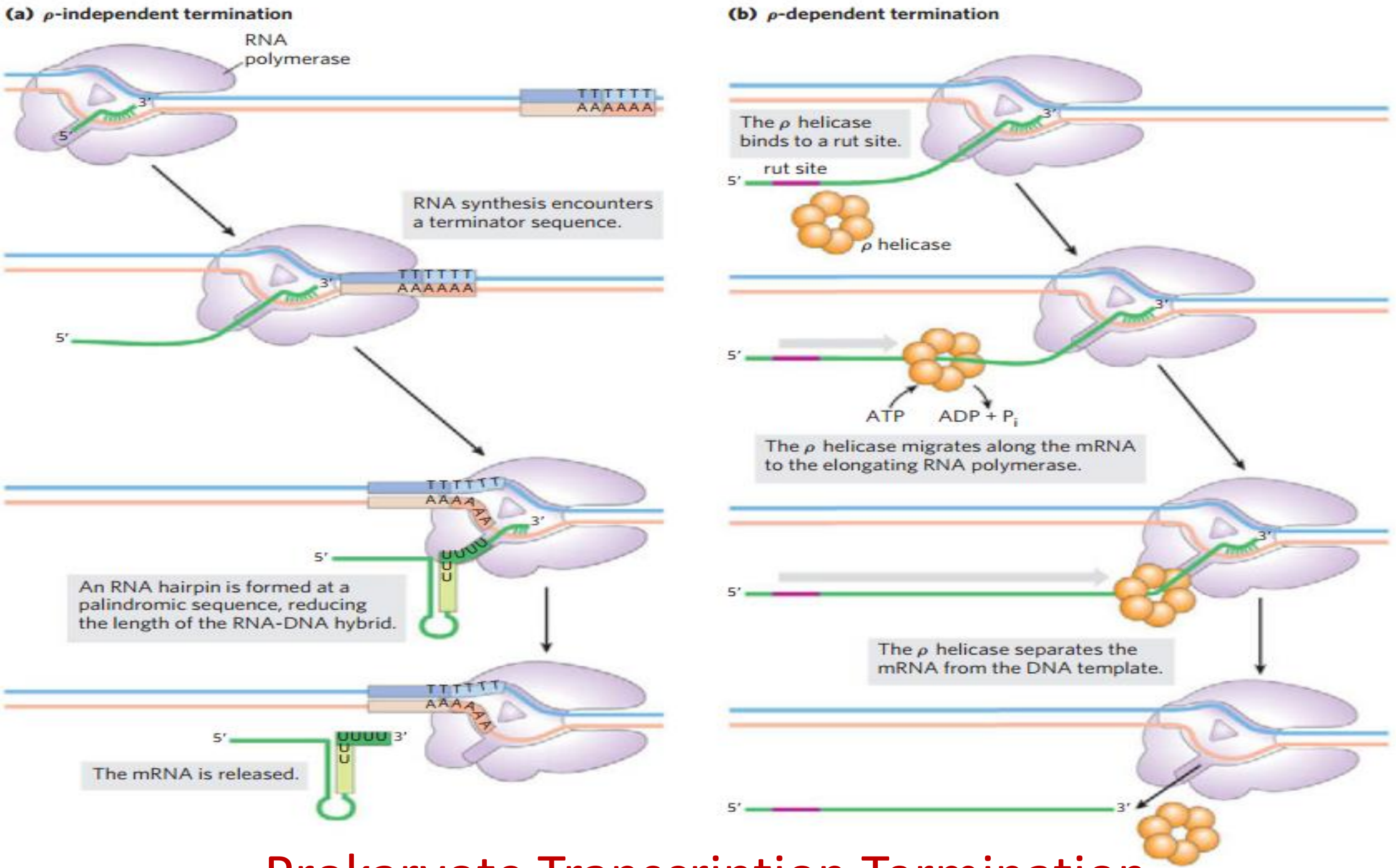
- DNA gồm 3 phần chính: promoter, trình tự mã hóa, và kết thúc phiên mã (terminator)



Prokaryote Transcription



1. RNA polymerase core binds to the DNA promoter.
2. Transcription bubble forms.
3. Transcription is initiated.
4. Promoter clearance is followed by elongation.
5. Elongation continues. σ^{70} dissociates, and is replaced by NusA.
6. Transcription is terminated. NusA dissociates, and the RNA polymerase is recycled.



RNA synthesis encounters a terminator sequence.

An RNA hairpin is formed at a palindromic sequence, reducing the length of the RNA-DNA hybrid.

The mRNA is released.

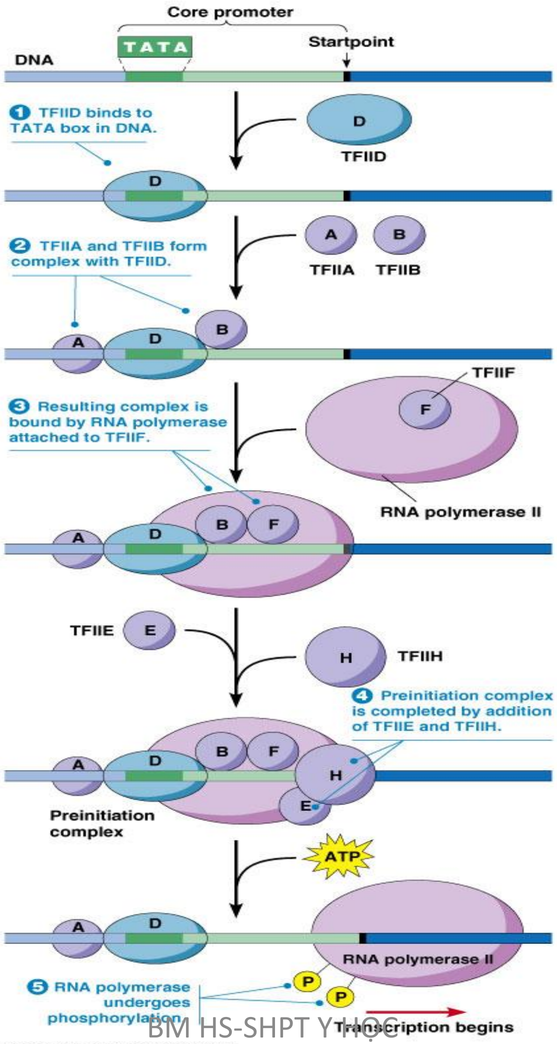
The ρ helicase binds to a rut site.

The ρ helicase migrates along the mRNA to the elongating RNA polymerase.

The ρ helicase separates the mRNA from the DNA template.

Prokaryote Transcription Termination

Eukaryote Transcription Initiation



❶ TFIID binds to TATA box in DNA.

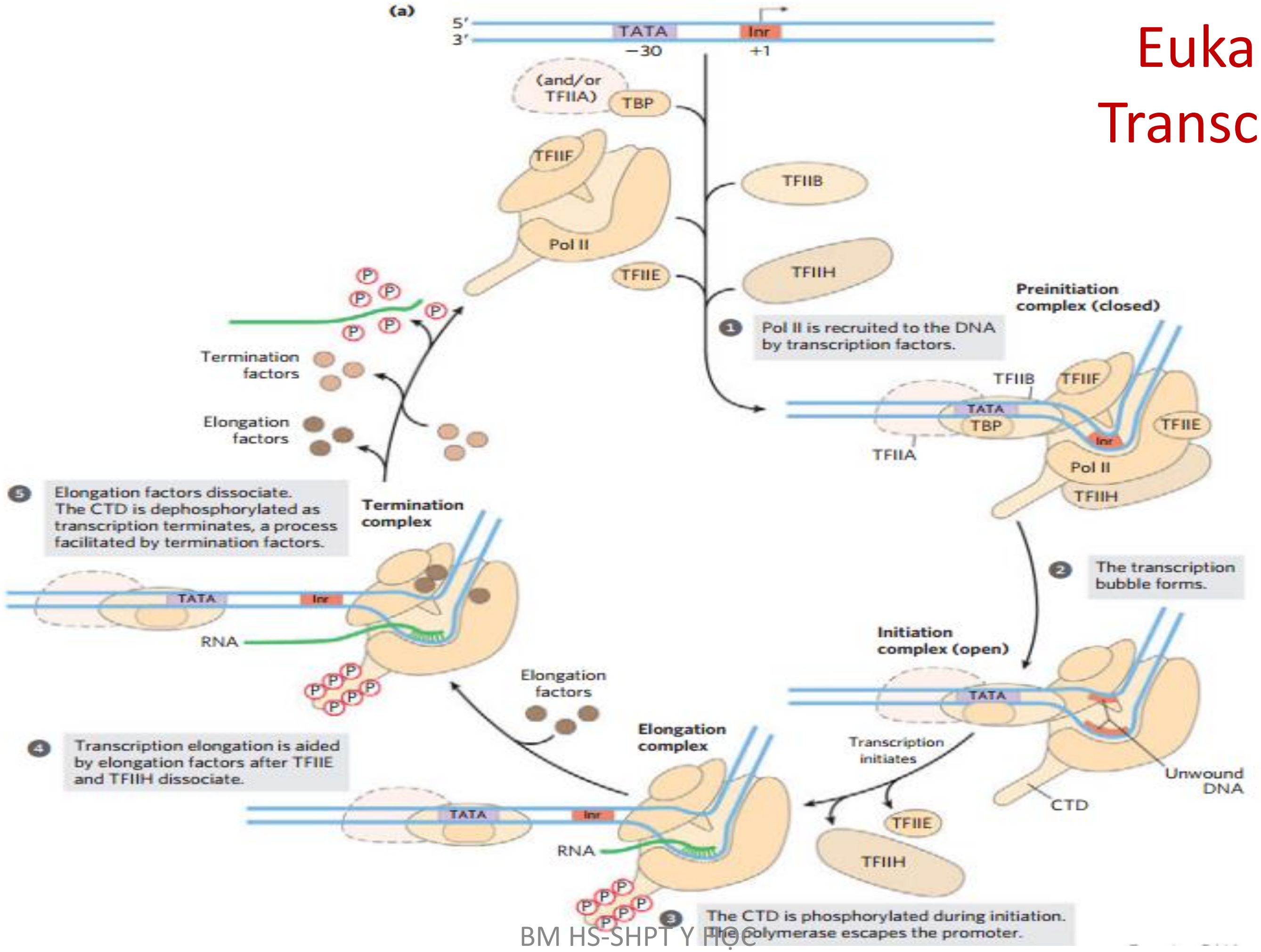
❷ TFIIA and TFIIB form complex with TFIID.

❸ Resulting complex is bound by RNA polymerase attached to TFIIF.

❹ Preinitiation complex is completed by addition of TFIIE and TFIIH.

❺ RNA polymerase undergoes phosphorylation.

Eukaryote Transcription



❶ Pol II is recruited to the DNA by transcription factors.

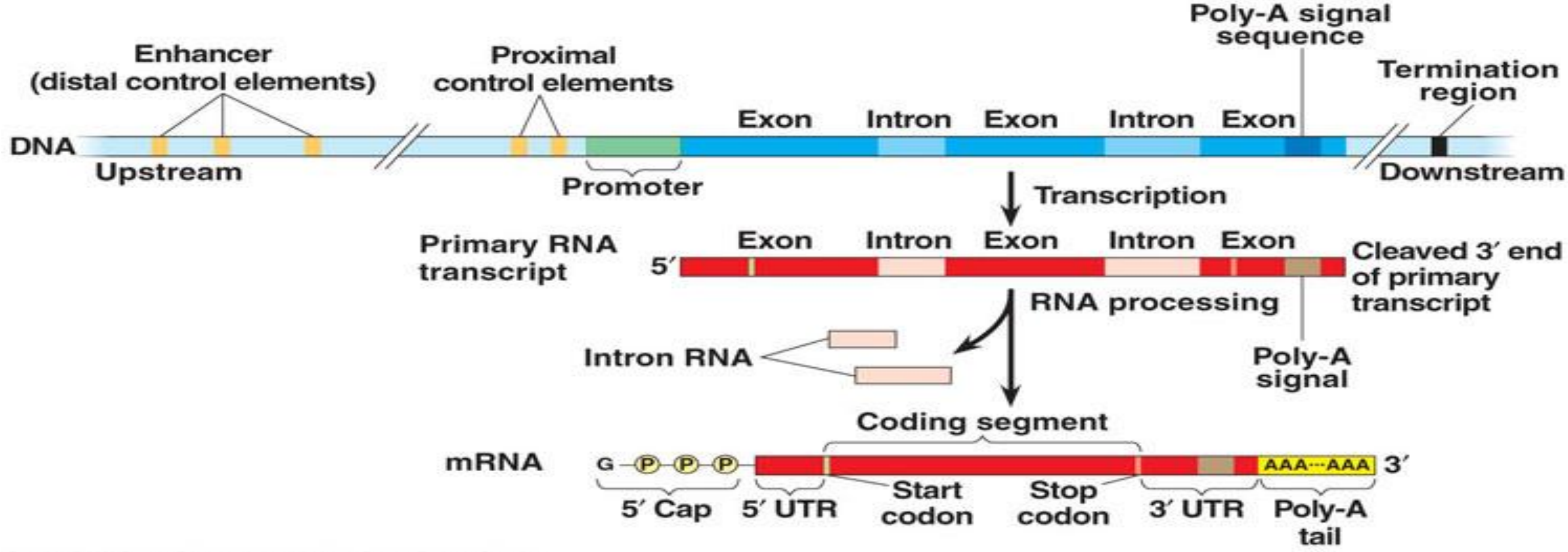
❷ The transcription bubble forms.

❸ The CTD is phosphorylated during initiation. The polymerase escapes the promoter.

❹ Transcription elongation is aided by elongation factors after TFIIE and TFIIH dissociate.

❺ Elongation factors dissociate. The CTD is dephosphorylated as transcription terminates, a process facilitated by termination factors.

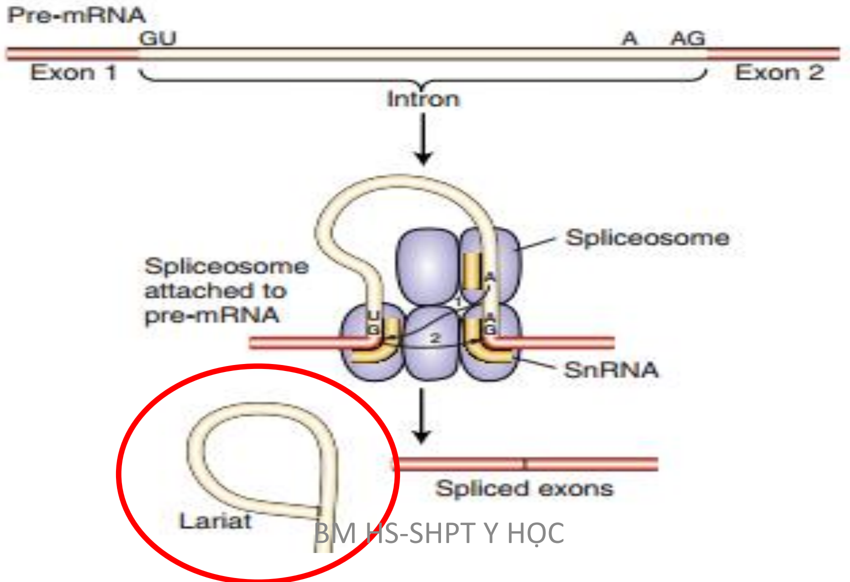
Quá trình trưởng thành của mRNA



Quá trình trưởng thành của mRNA

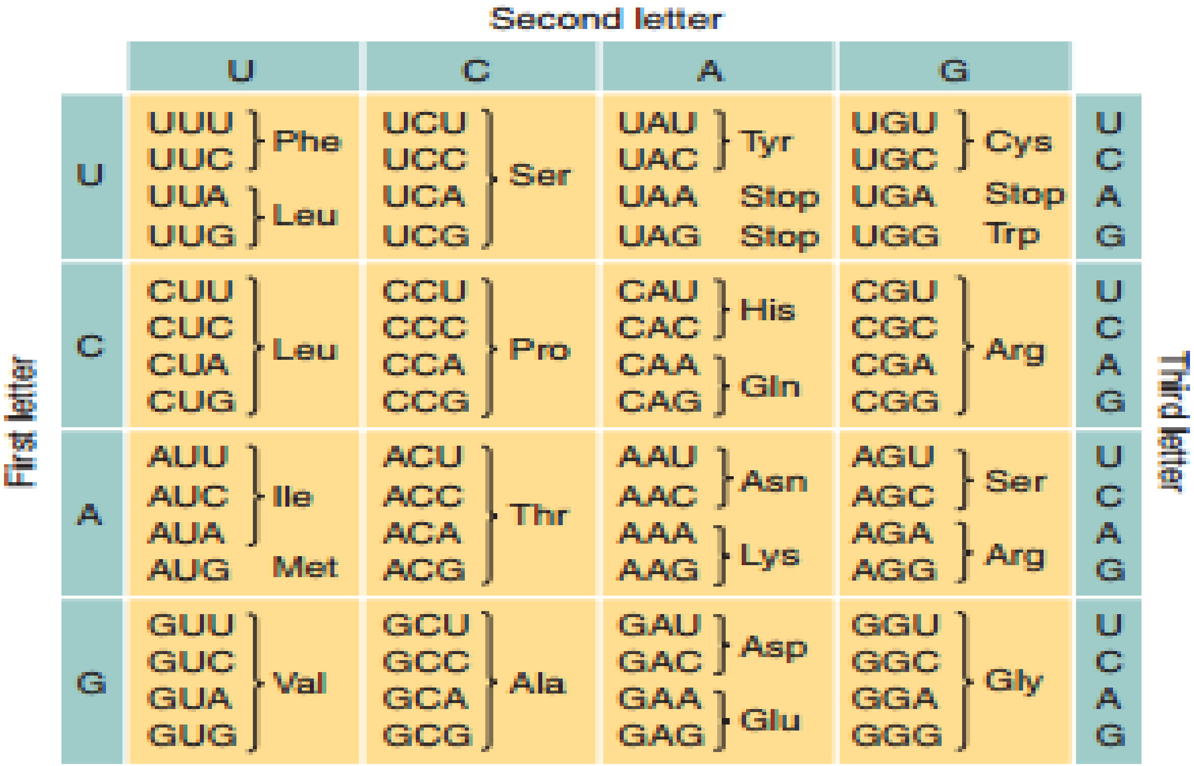
Quá trình cắt nối pre mRNA – splicing

- Cắt bỏ các đoạn intron và nối các đoạn exon
- Enzyme là splicesome (thể cắt nối)



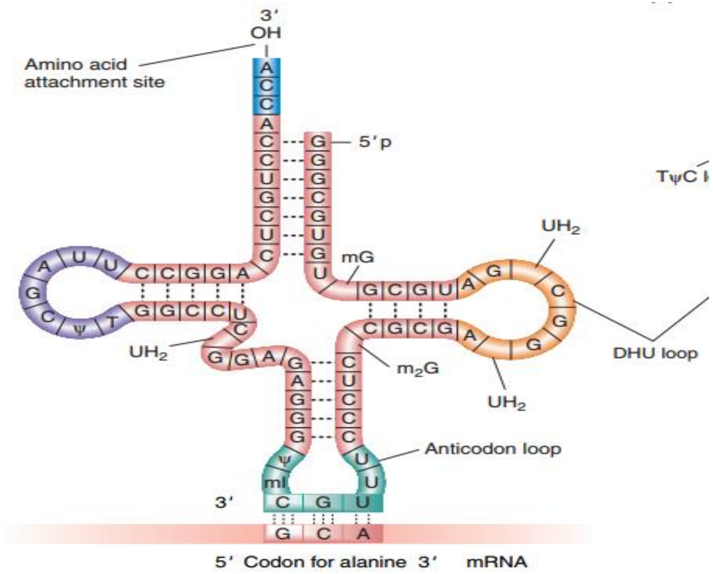
TRANSLATION

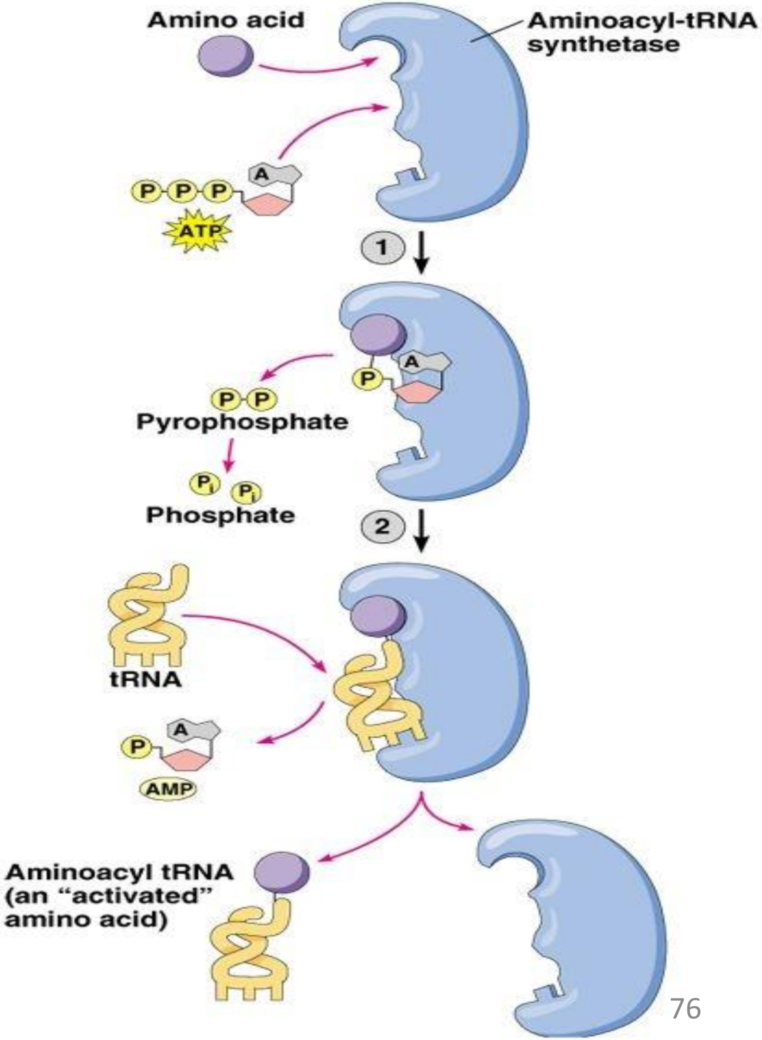
Mã di truyền



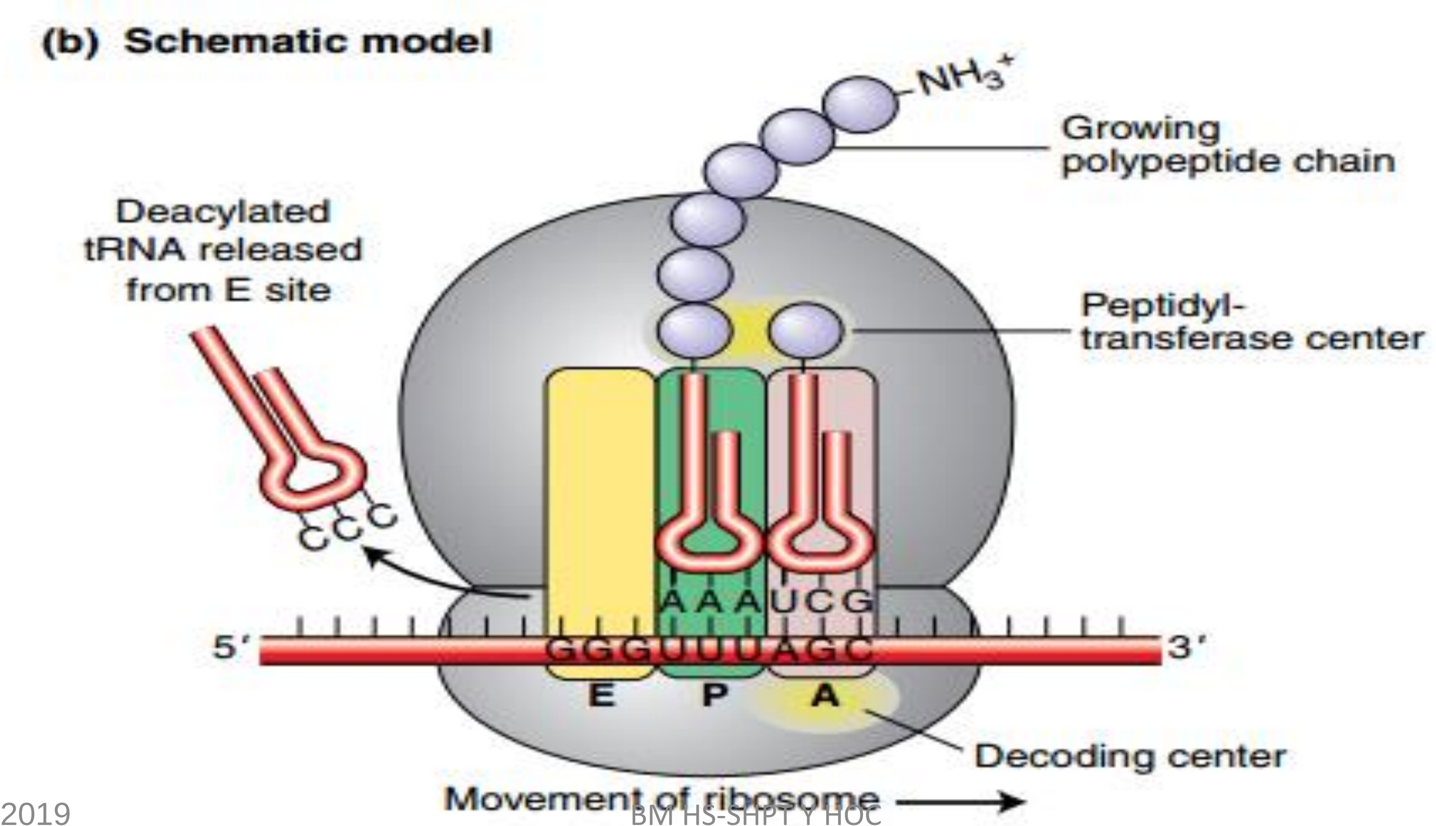
First letter	Second letter								Third letter
	U		C		A		G		
U	UUU	Phe	UCU	Ser	UAU	Tyr	UGU	Cys	U
	UUC		UCC		UAC		UGC		C
	UUA	Leu	UCA		UAA	Stop	UGA	Stop	A
	UUG		UCG		UAG	Stop	UGG	Trp	G
C	CUU	Leu	CCU	Pro	CAU	His	CGU	Arg	U
	CUC		CCC		CAC		CGC		C
	CUA		CCA		CAA	Gln	CGA		A
	CUG		CCG		CAG		CGG		G
A	AUU	Ile	ACU	Thr	AAU	Asn	AGU	Ser	U
	AUC		ACC		AAC		AGC		C
	AUA		ACA		AAA	Lys	AGA	Arg	A
	AUG	Met	ACG		AAG		AGG		G
G	GUU	Val	GCU	Ala	GAU	Asp	GGU	Gly	U
	GUC		GCC		GAC		GGC		C
	GUA		GCA		GAA	Glu	GGA		A
	GUG		GCG		GAG		GGG		G

tRNA


3′
OH
Amino acid attachment site
5′p
TψC l
UH₂
mG
DHU loop
m₂G
UH₂
UH₂
Anticodon loop
mI
ψ
3′
C G U
G C A
5′ Codon for alanine 3′
mRNA


Amino acid
Aminoacyl-tRNA synthetase
P P P
A
ATP
1
P
Pyrophosphate
Pᵢ Pᵢ
Phosphate
2
tRNA
AMP
Aminoacyl tRNA (an "activated" amino acid)

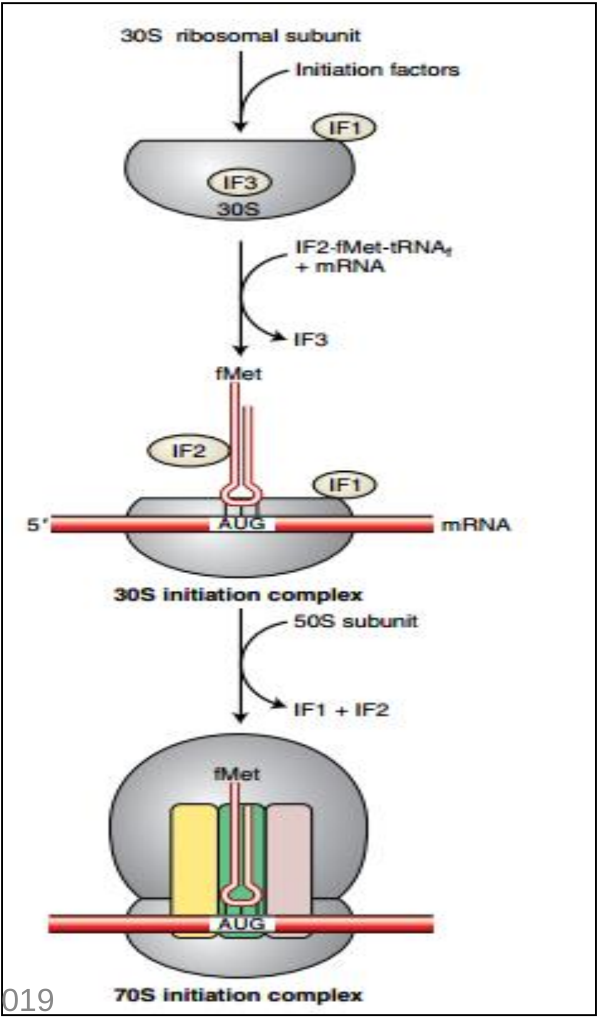
Ribosome

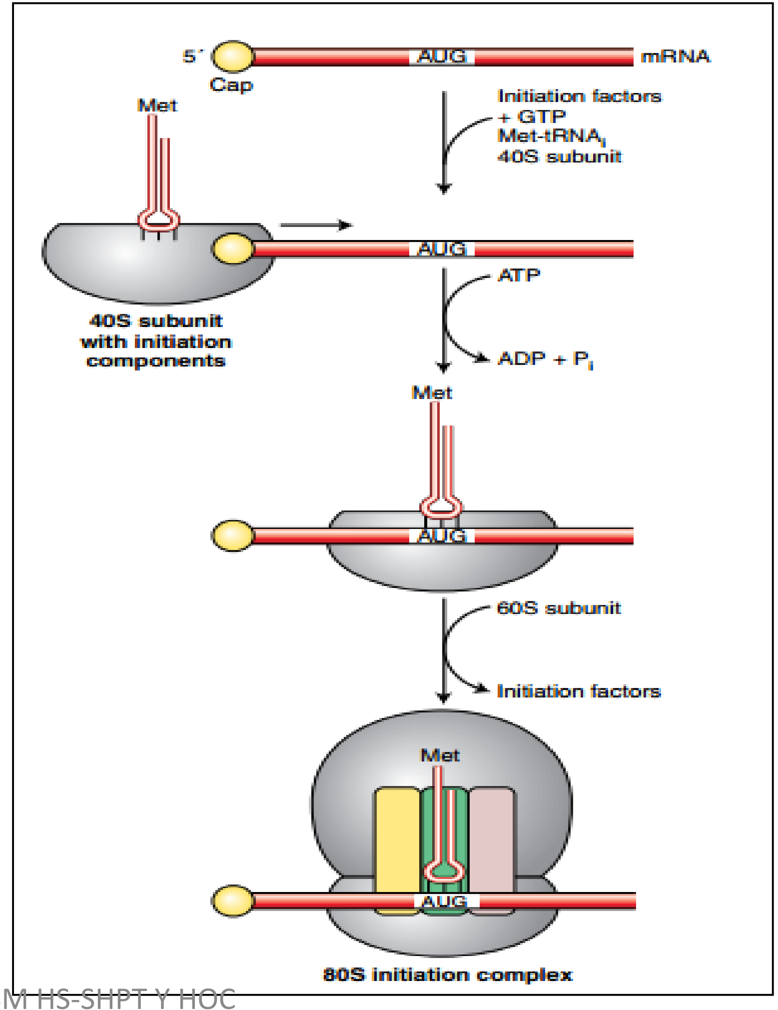

(b) Schematic model
NH3+
Growing polypeptide chain
Deacylated tRNA released from E site
Peptidyl-transferase center
CCC
AAAUCG
5′
GGGUUUAGC
3′
E
P
A
Decoding center
Movement of ribosome

Quá trình dịch mã

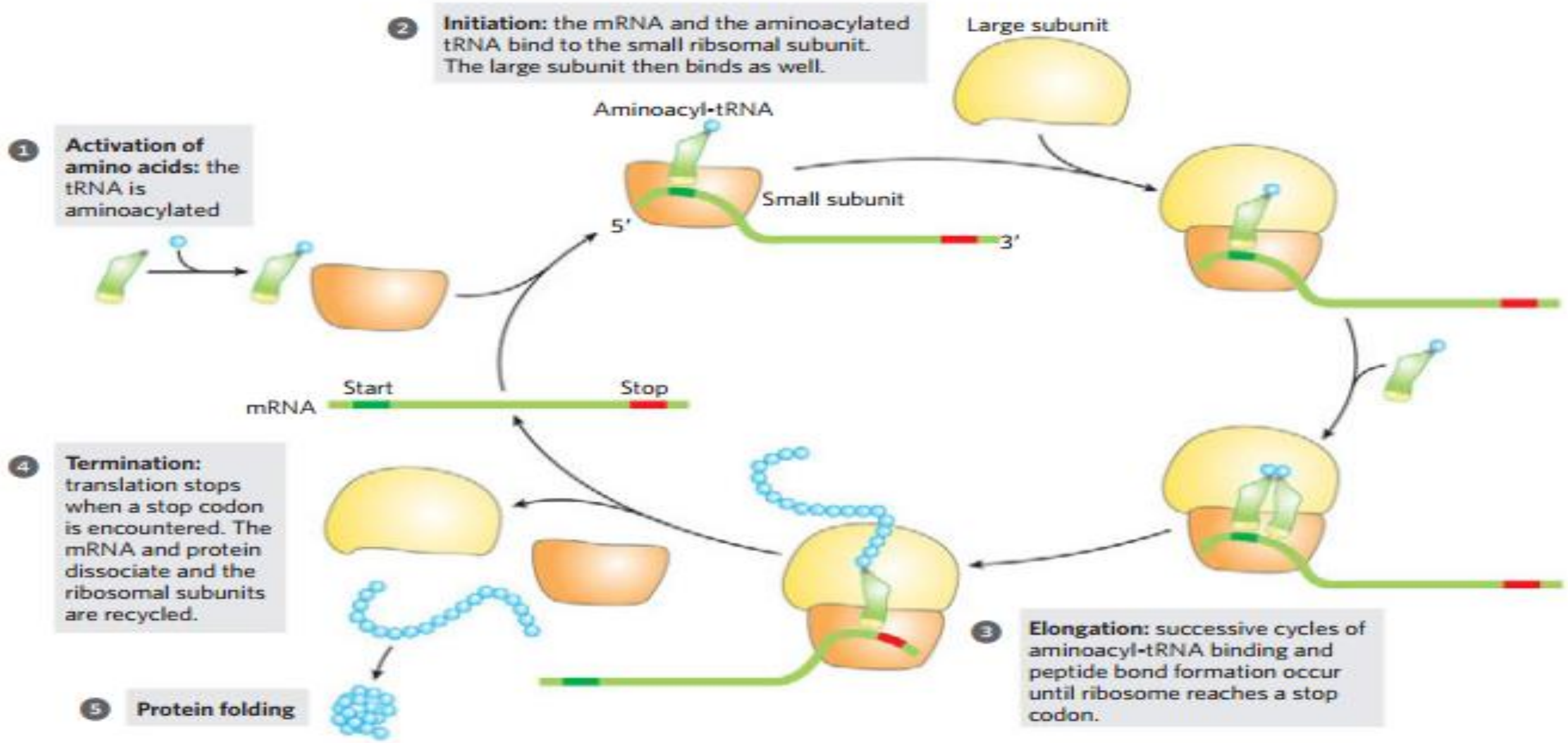
- Không có sự khác biệt lớn trong quá trình dịch mã giữa prokaryote và eukaryote, trừ giai đoạn mở đầu
    i. giai đoạn mở đầu (initiation)
    ii. giai đoạn kéo dài (elongation)
    iii. giai đoạn kết thúc (termination)

prokaryote





eukaryote



❶ **Activation of amino acids:** the tRNA is aminoacylated

❷ **Initiation:** the mRNA and the aminoacylated tRNA bind to the small ribsomal subunit. The large subunit then binds as well.

❸ **Elongation:** successive cycles of aminoacyl-tRNA binding and peptide bond formation occur until ribosome reaches a stop codon.

❹ **Termination:** translation stops when a stop codon is encountered. The mRNA and protein dissociate and the ribosomal subunits are recycled.

❺ **Protein folding**

IN SUMMARY

- Gene là gì? Genome là gì?
- Có bao nhiêu loại gene?
- Cơ thể có cơ chế gì để đảm bảo số lượng gene phù hợp với lượng chức năng nhiệm vụ nó bảo đảm?
- Khác nhau giống nhau DNA và RNA? Tại sao?
- Chức năng nhiệm vụ của DNA và RNA? Tại sao?
- Giai đoạn nào của các quá trình truyền tải thông tin di truyền là quan trọng nhất? Tại sao? Có sự giống nhau giữa các tế bào prokaryote và eukaryote không? Nếu có thì như thế nào? Nếu không thì tại sao?

Tài liệu tham khảo

- Pray, L. (2008) *Transposons, or jumping genes: Not junk DNA?* Nature Education 1(1):32
- Griffiths, A. J. (2005). *An introduction to genetic analysis.* Macmillan.
- Reece, J. B., Urry, L. A., Cain, M. L., Wasserman, S. A., Minorsky, P. V., & Jackson, R. (2014). *Campbell biology* (p. 135). Boston: Pearson.
- Nelson, D. L., Lehninger, A. L., & Cox, M. M. (2008). *Lehninger principles of biochemistry*. Macmillan.

Any question?

THANK YOU