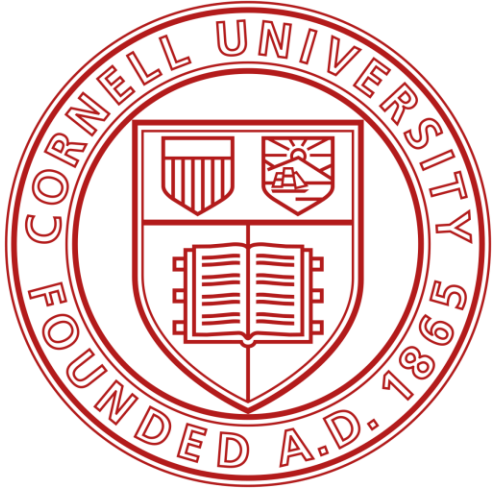


LUMIERE EDUCATION

Lumiere Education Biochemistry: DNA, Proteins, and their Applications Lecture 1

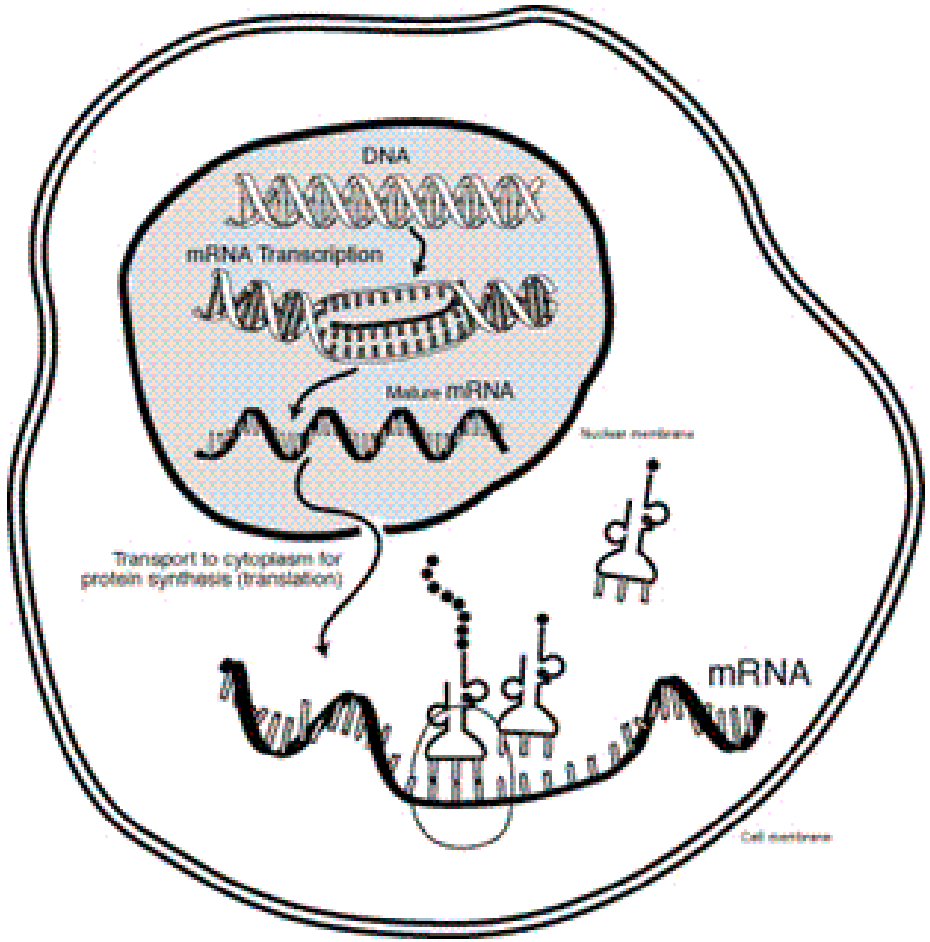
Introductions & Transcription, Translation, and Protein QC

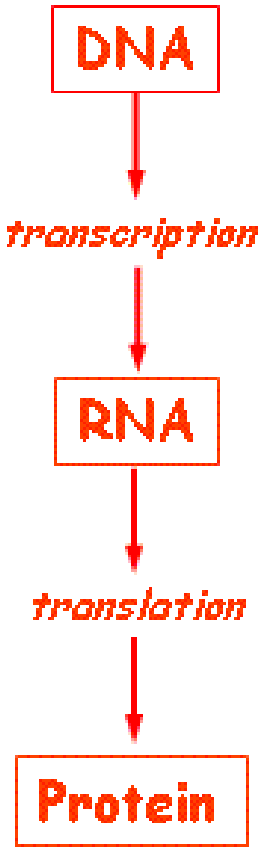
Ari Broad

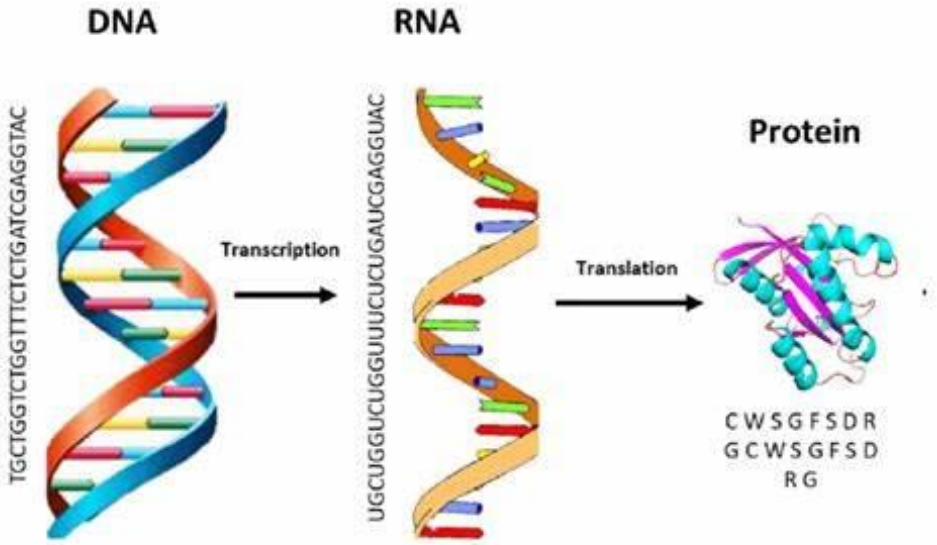
Weill Institute for Cell and Molecular Biology (WICMB)

Cornell University

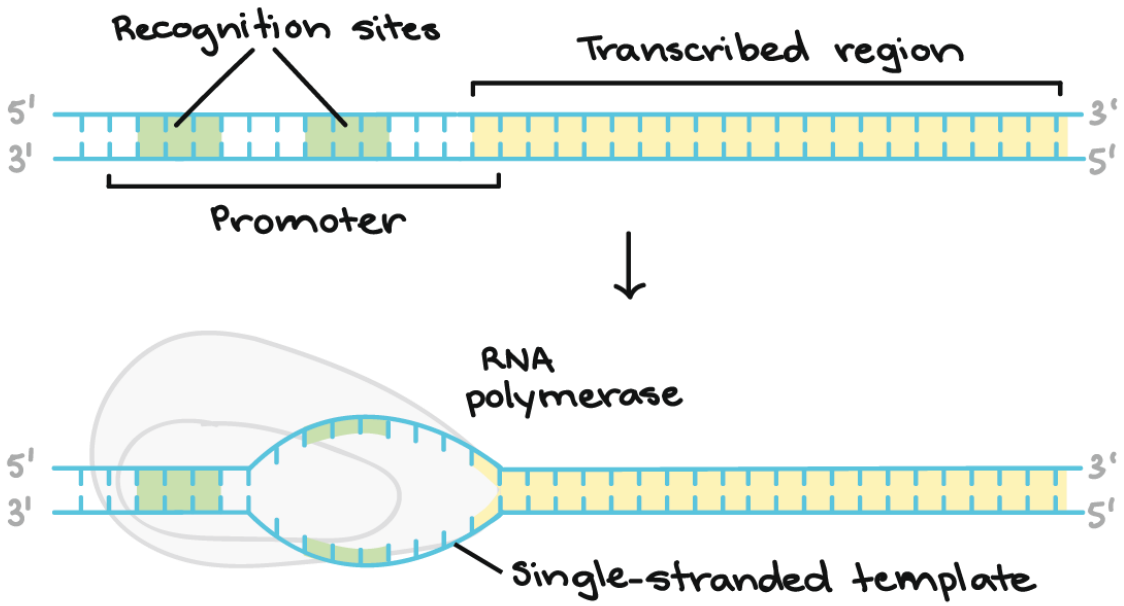
Central Dogma Review

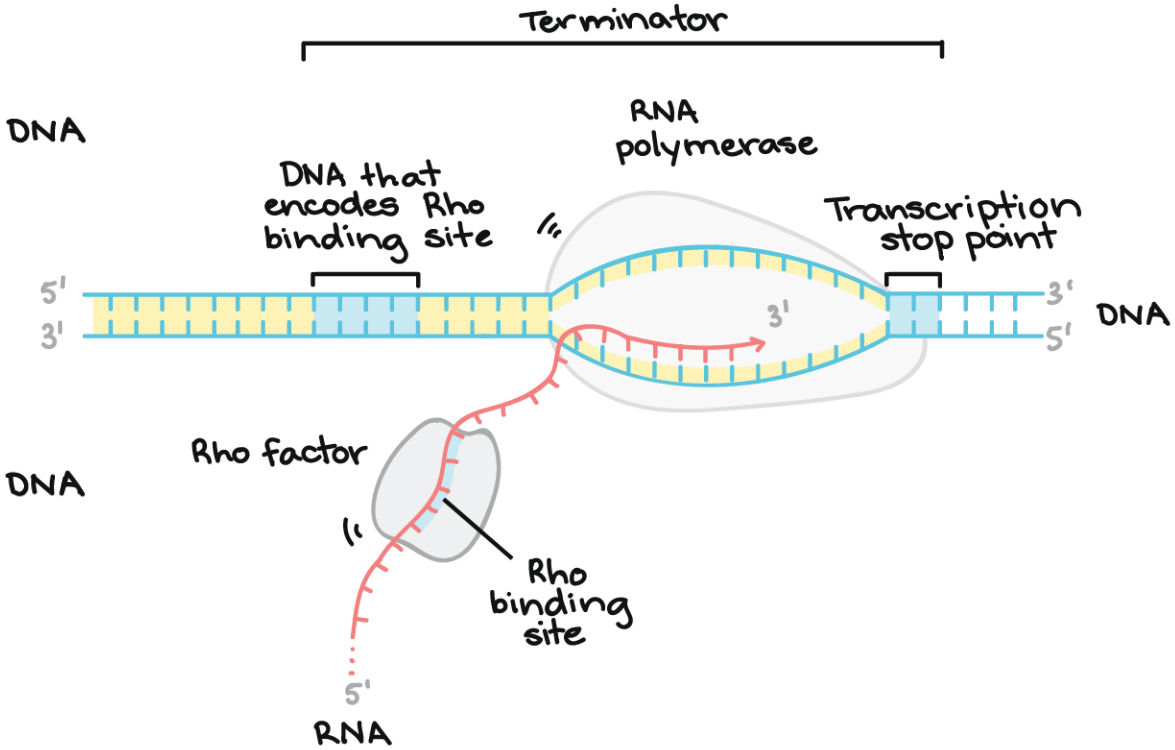






Transcription Review

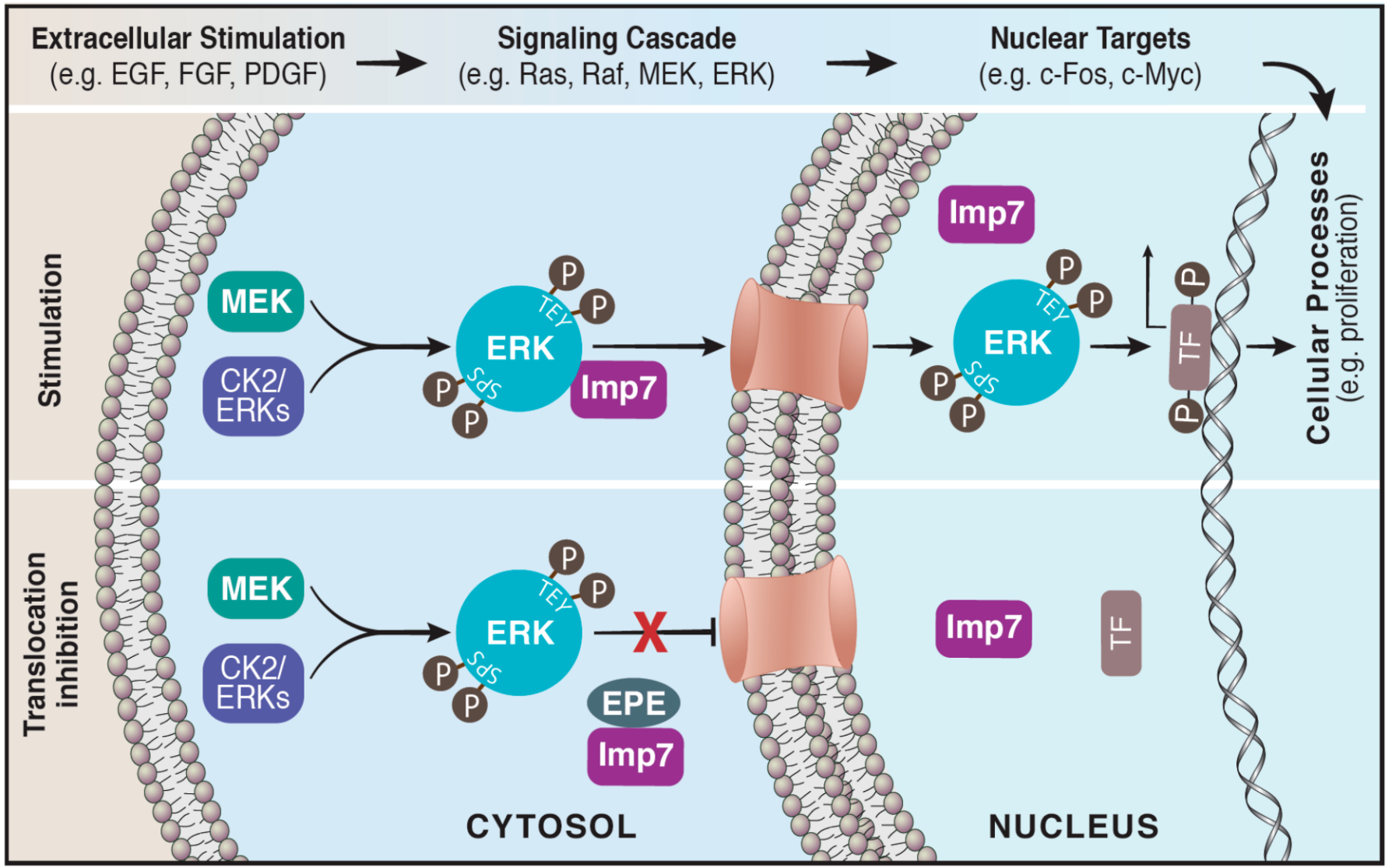




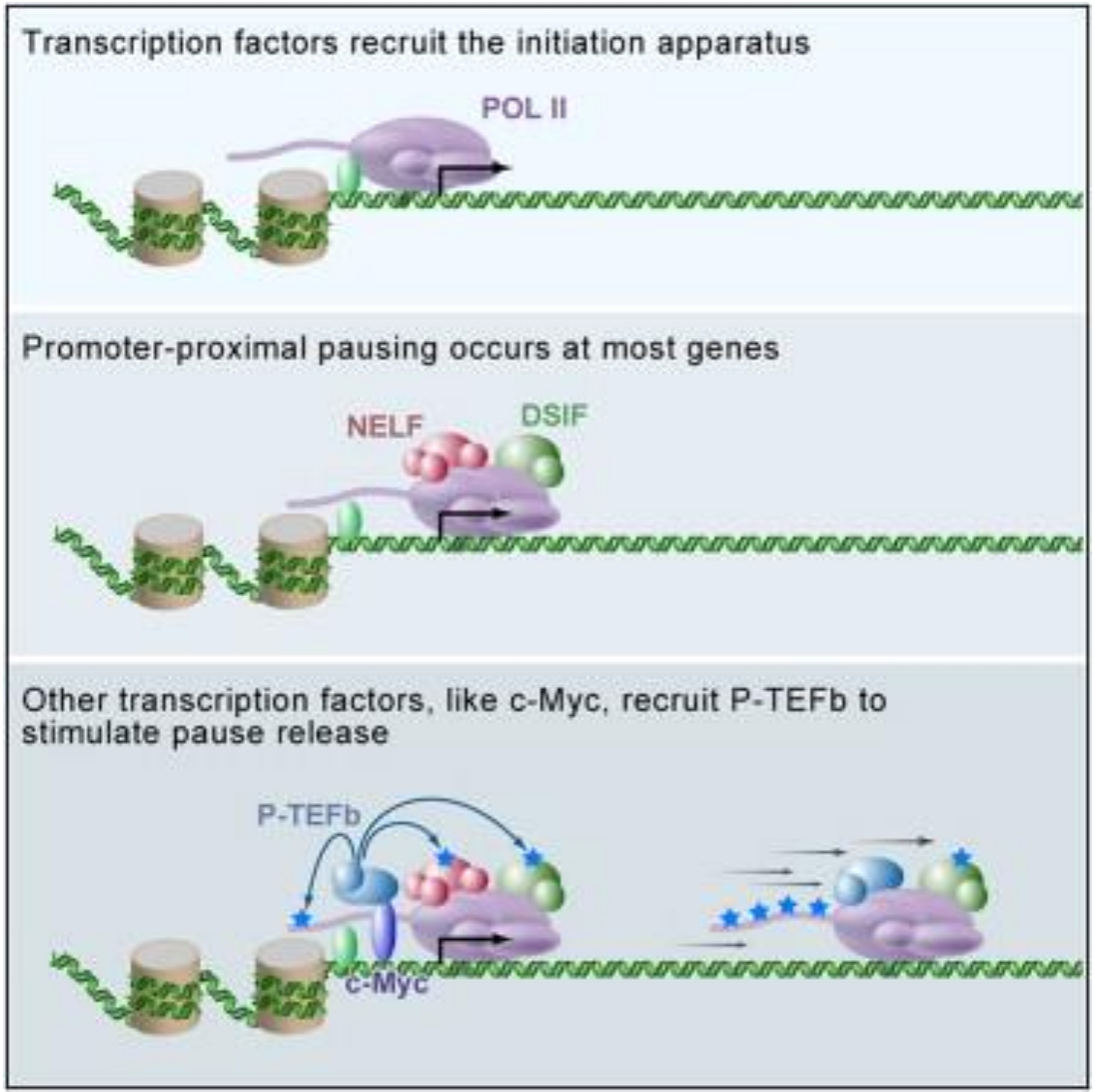
Transcription as a Field – What are the standing questions?

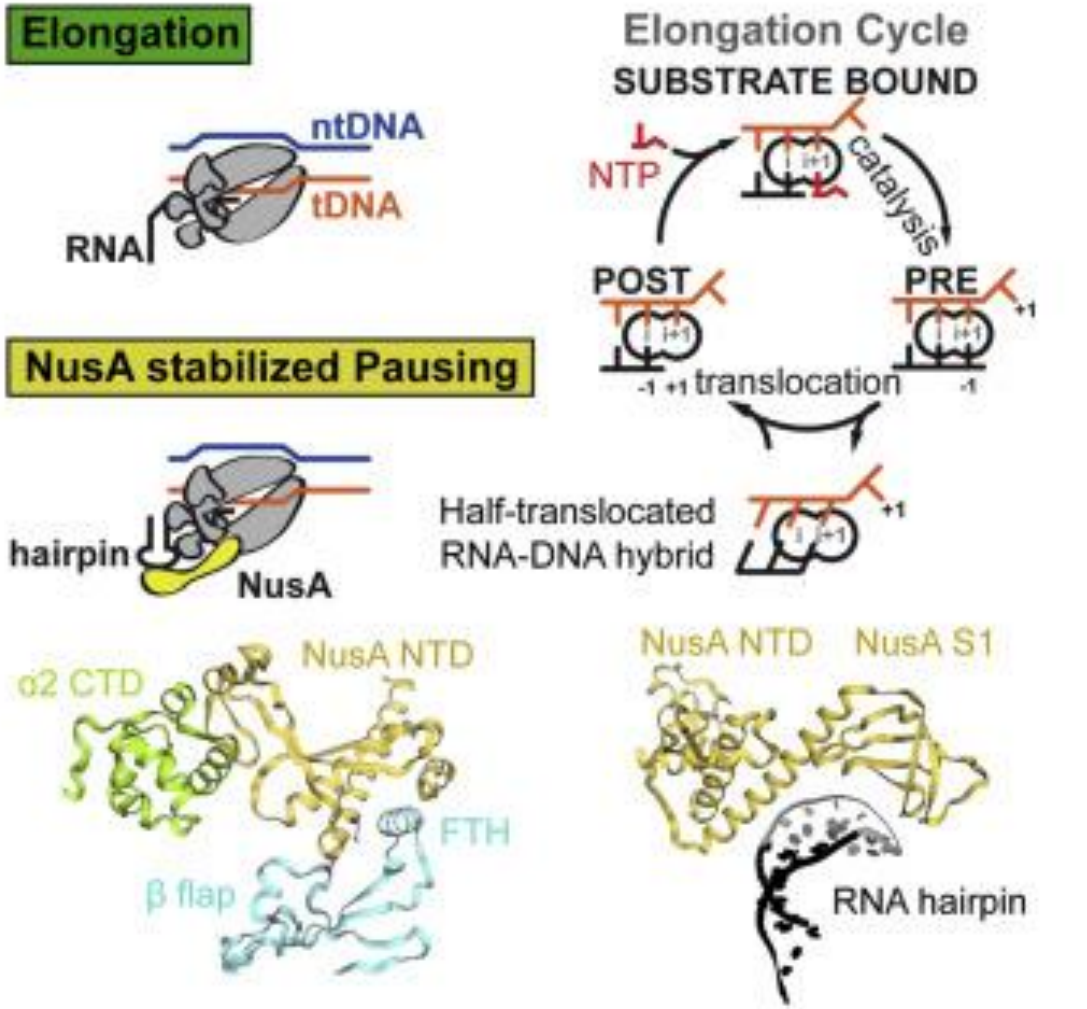
- A LOT…
- There is new research coming out every day discovering different proteins that translocate to the nucleus to aide in transcription activation and repression.
- But a field that is the "hottest" in transcription is about the transcriptional regulation of pausing.
- There are other transcription fields that are emerging such as transposable elements.

Transcription factors translocating to the nucleus



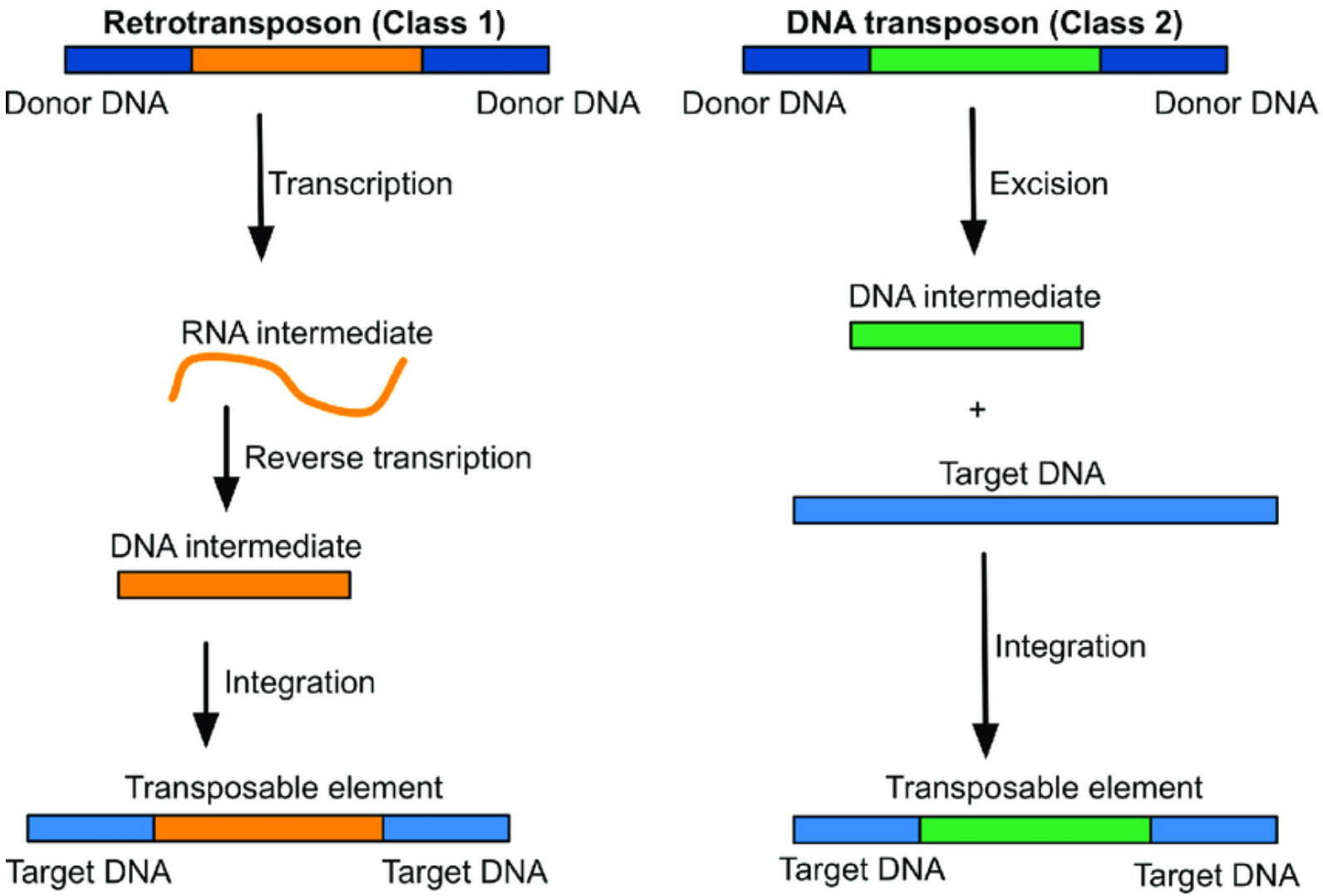
Transcriptional Pausing



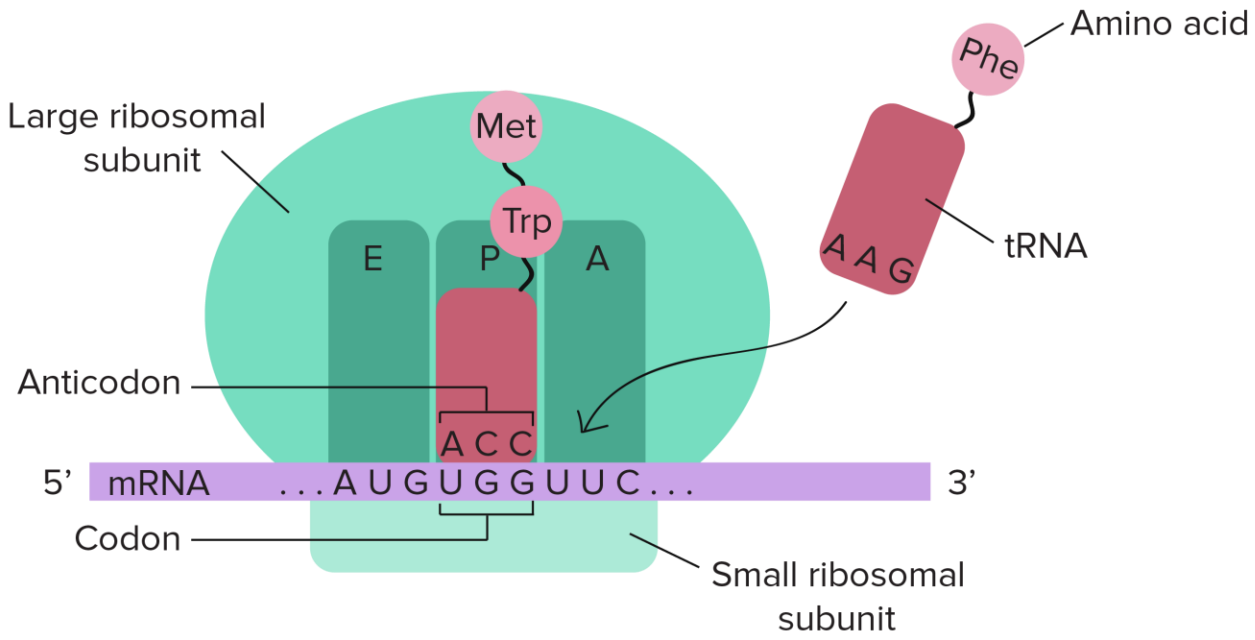


NusA stabilizes paused RNAP elongation complex through interactions with RNAP and creating a positively charged cavity above the RNA hairpin

Transposable Elements



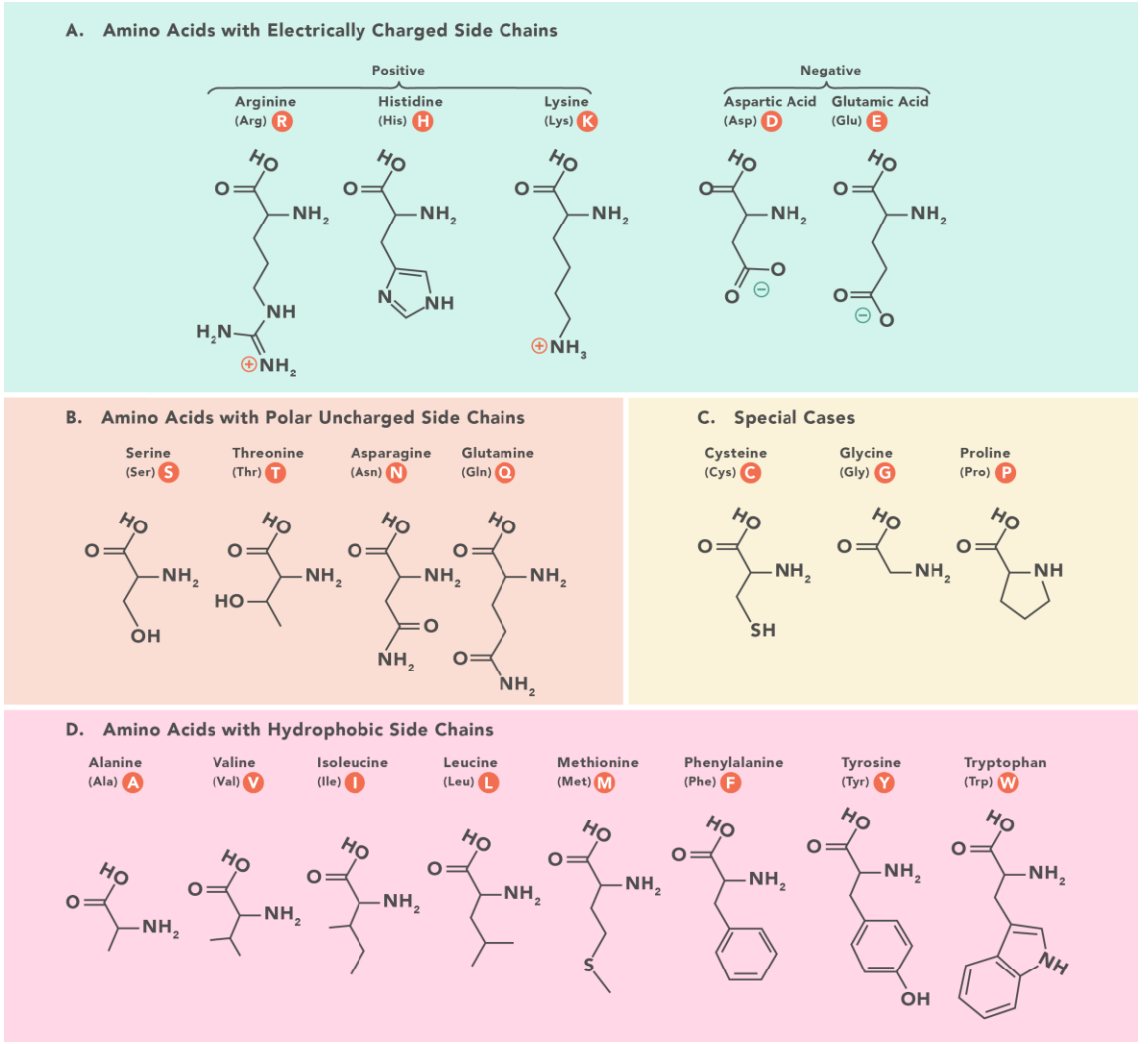
Translation Review



Second Position

First Position (5' end)	U	C	A	G	Third Position (3' end)
U	UUU Phe UUC UUA Leu UUG	UCU UCC UCA Ser UCG	UAU Tyr UAC UAA Stop UAG Stop	UGU Cys UGC UGA Stop UGG Trp	U C A G
C	CUU CUC CUA Leu CUG	CCU CCC CCA Pro CCG	CAU His CAC CAA Gln CAG	CGU CGC CGA Arg CGG	U C A G
A	AUU AUC Ile AUA AUG Met	ACU ACC ACA Thr ACG	AAU Asn AAC AAA Lys AAG	AGU Ser AGC AGA Arg AGG	U C A G
G	GUU GUC GUA Val GUG	GCU GCC GCA Ala GCG	GAU Asp GAC GAA Glu GAG	GGU GGC GGA Gly GGG	U C A G

Amino Acids

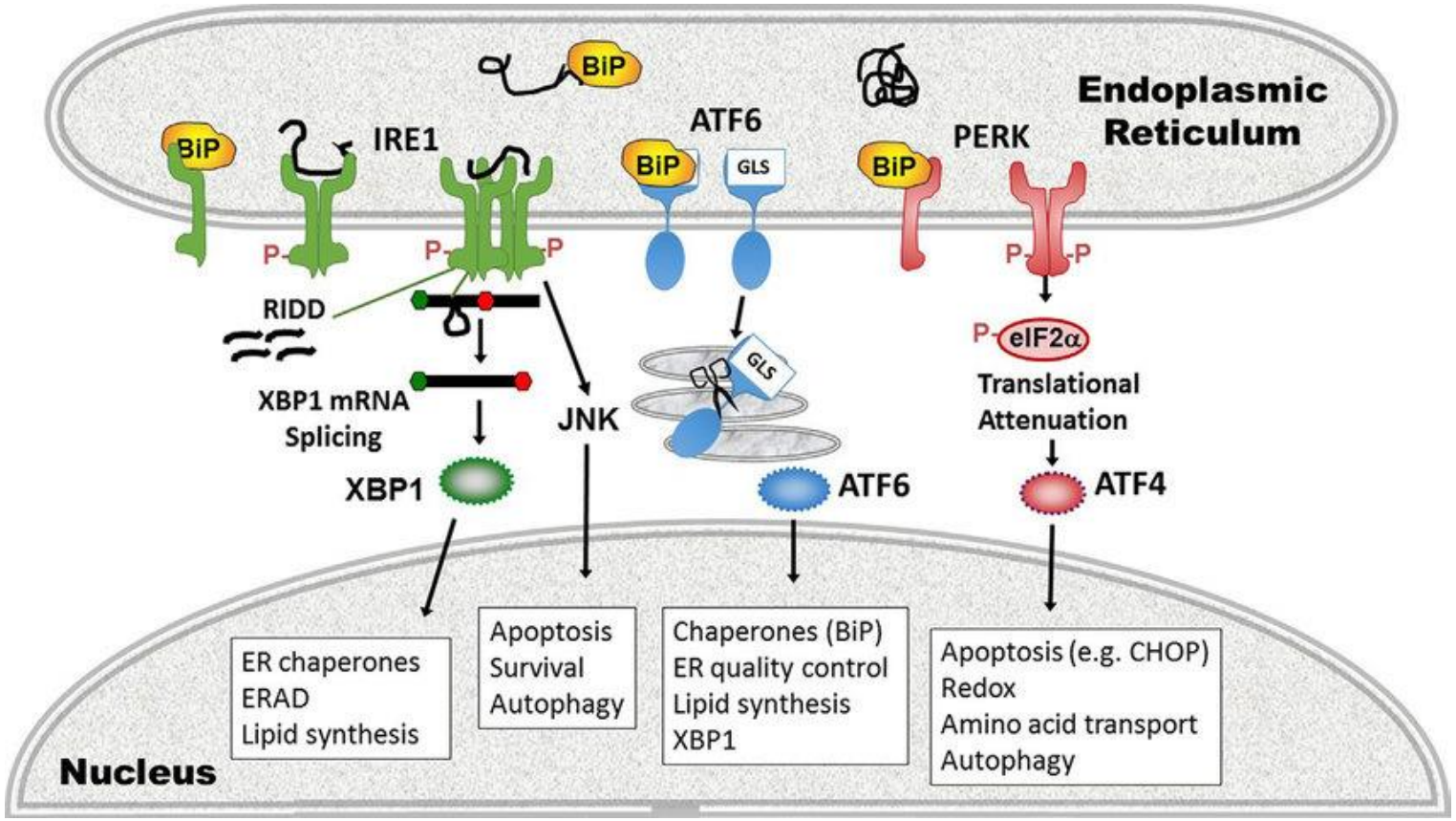

A. Amino Acids with Electrically Charged Side Chains
Positive
Arginine (Arg) R
Histidine (His) H
Lysine (Lys) K
Negative
Aspartic Acid (Asp) D
Glutamic Acid (Glu) E
B. Amino Acids with Polar Uncharged Side Chains
Serine (Ser) S
Threonine (Thr) T
Asparagine (Asn) N
Glutamine (Gln) Q
C. Special Cases
Cysteine (Cys) C
Glycine (Gly) G
Proline (Pro) P
D. Amino Acids with Hydrophobic Side Chains
Alanine (Ala) A
Valine (Val) V
Isoleucine (Ile) I
Leucine (Leu) L
Methionine (Met) M
Phenylalanine (Phe) F
Tyrosine (Tyr) Y
Tryptophan (Trp) W

Translation as a Field – What are the standing questions?

- How is translation and nascent protein folding regulated?
- What are the mechanisms that protect nascent proteins during translation?

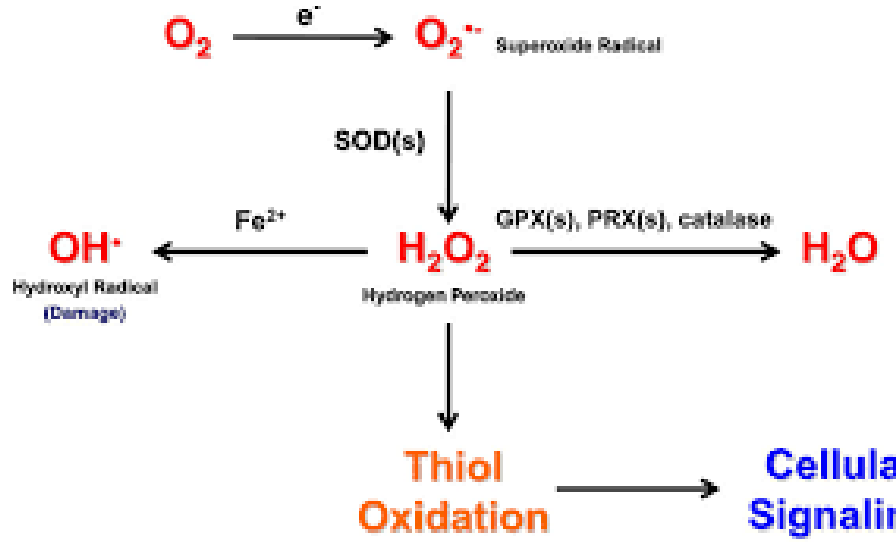
- This can be lumped into a field called Protein Quality Control (PQC).

Protein Quality Control – aka Unfolded Protein Response



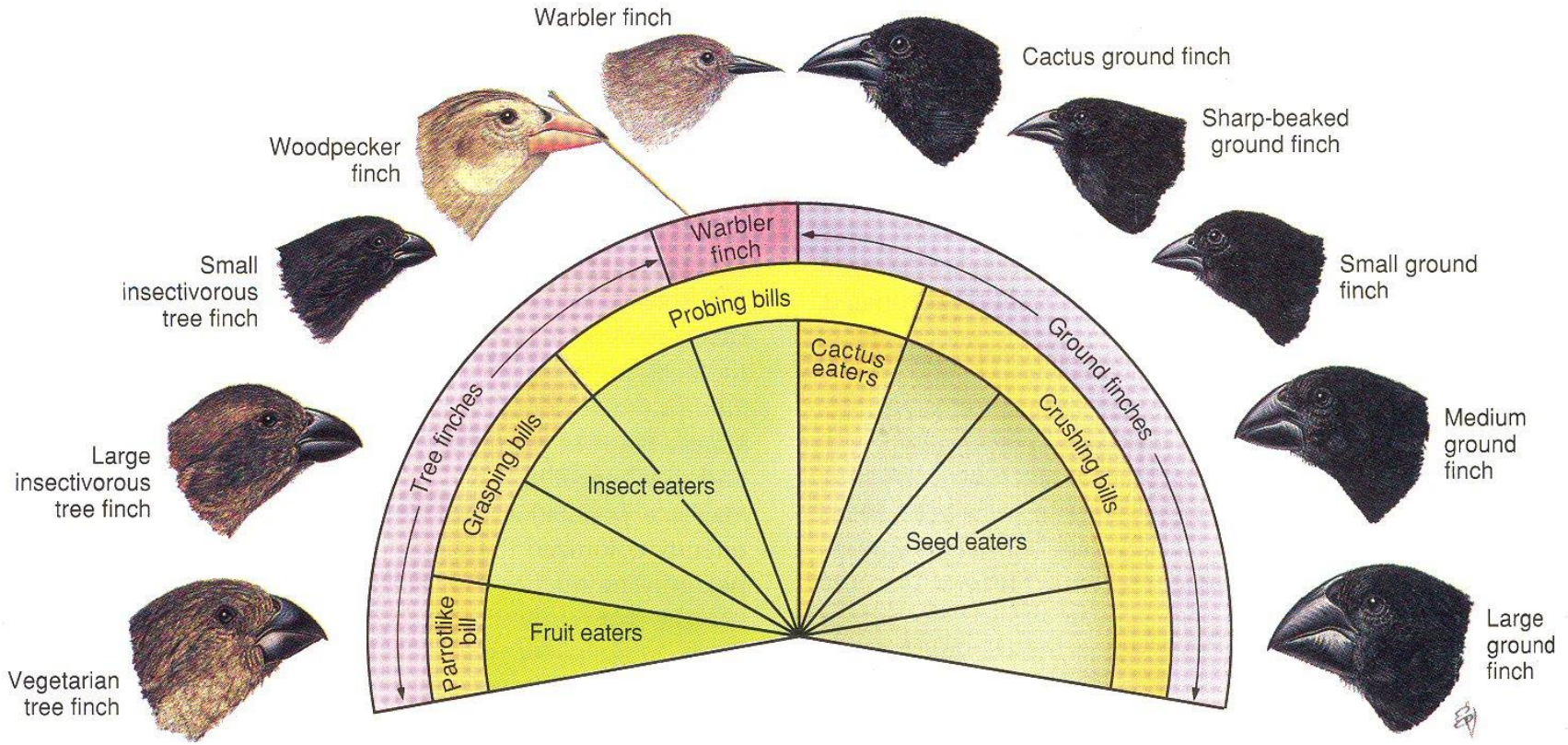
What can cause the UPR to be activated?

- Redox stress
- Osmotic stress
- Pathogen immune response



Protein Structure and Function

Structure is directly related to function

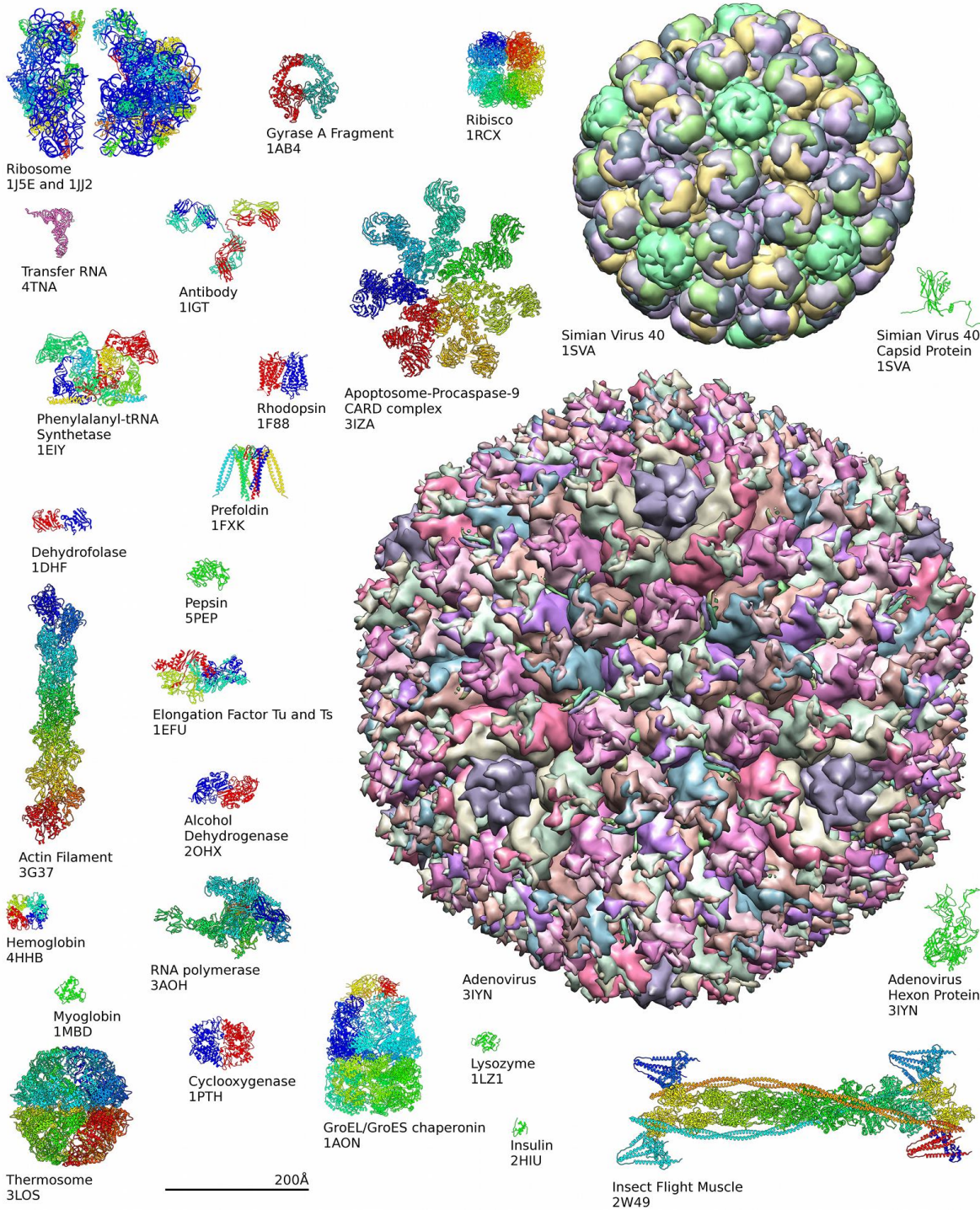


Protein Structure and Function

Structure is directly related to function

Protein Structure(Summary)

•Primary	The amino acid sequence	Glu-Arg-Phe-Gly
•Secondary	Characteristic structures that occur in many proteins (E.g. alpha helix , beta sheets)	alpha helix beta sheets
•Tertiary	Three dimensional structure of proteins	Y Z X
•Quaternary	Three dimensional structure of proteins composed of multiple subunits	

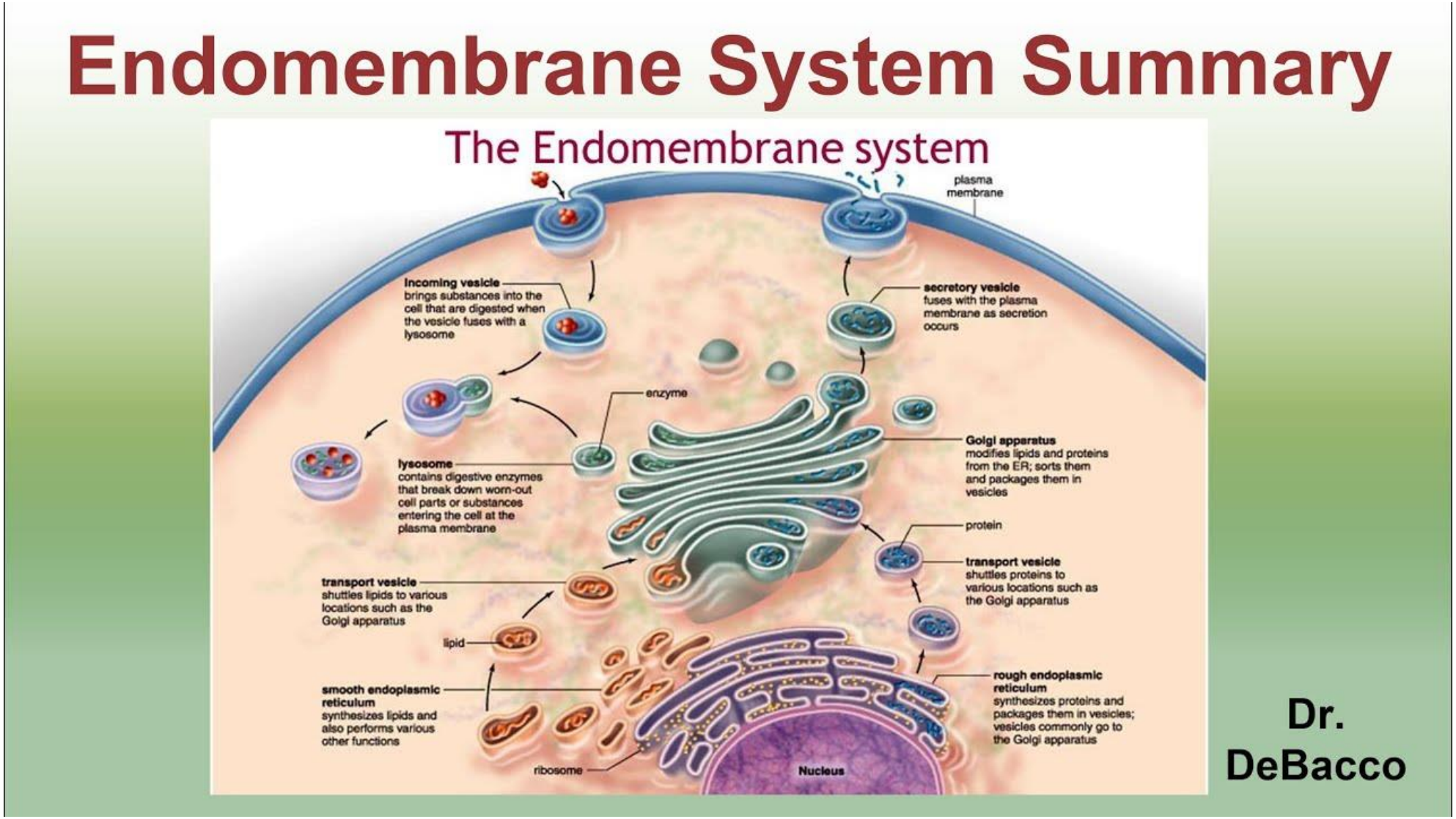


Summary

You should now be familiar with the following topics:

- Central Dogma
- Transcription and Transcriptional Regulation
- Translation
- Protein Quality Control
- Structure and Function

What I Do: The Golgi Apparatus



What I Do: Protein Structure and Function

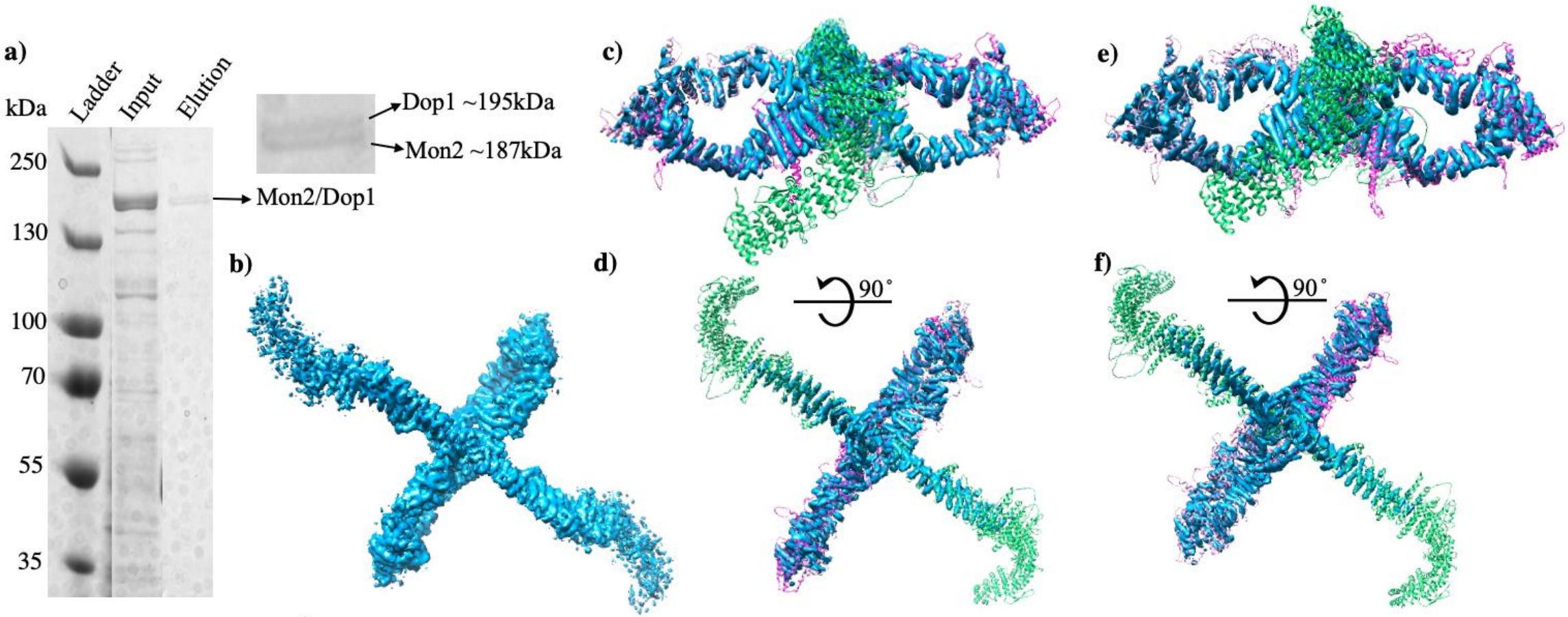
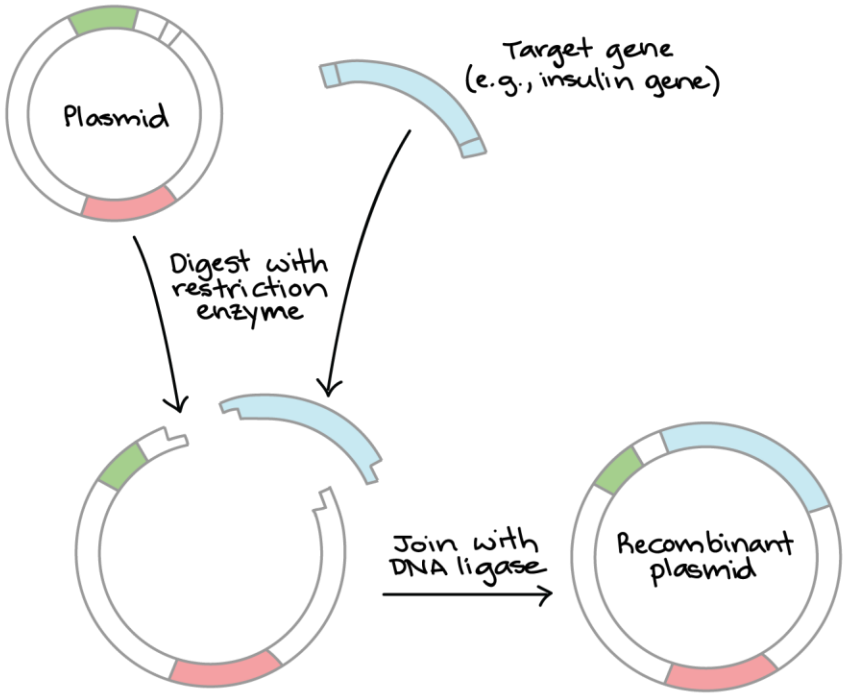
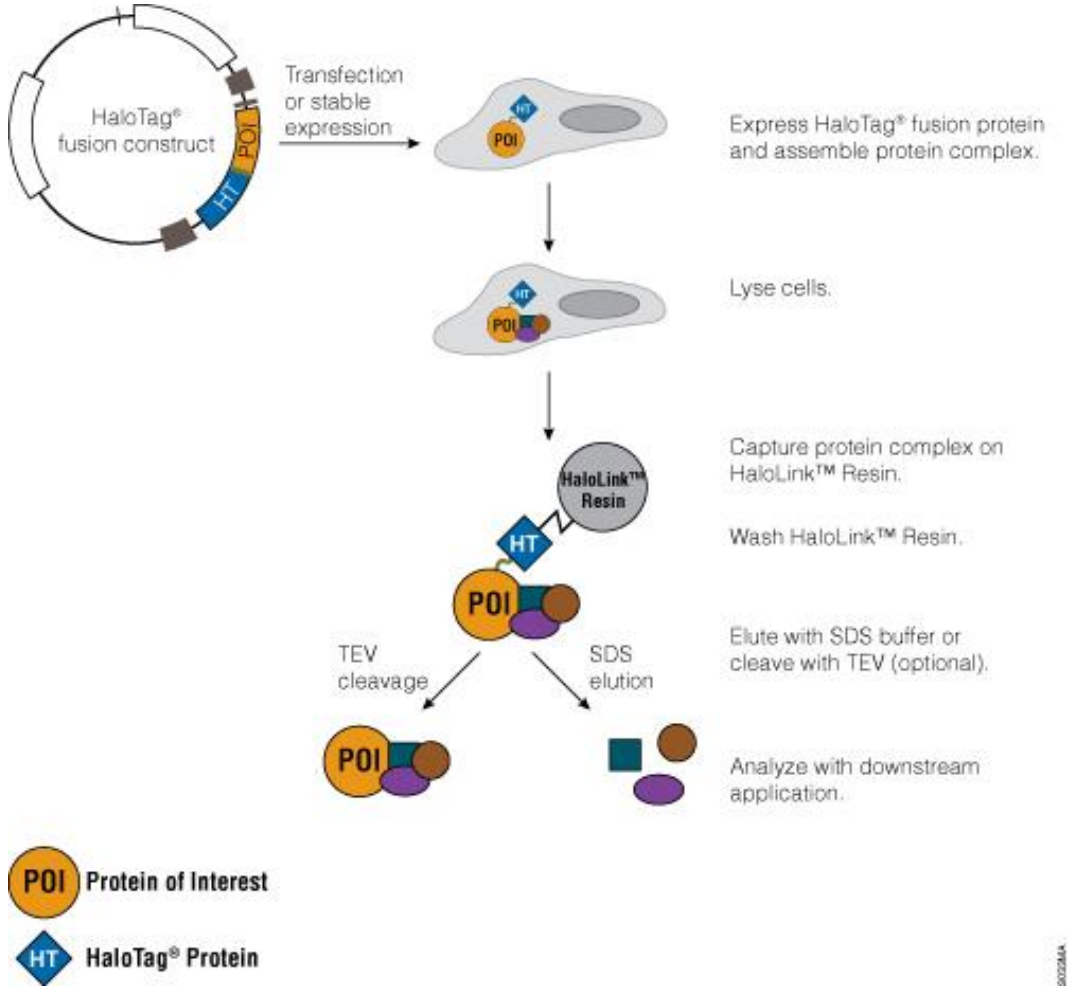


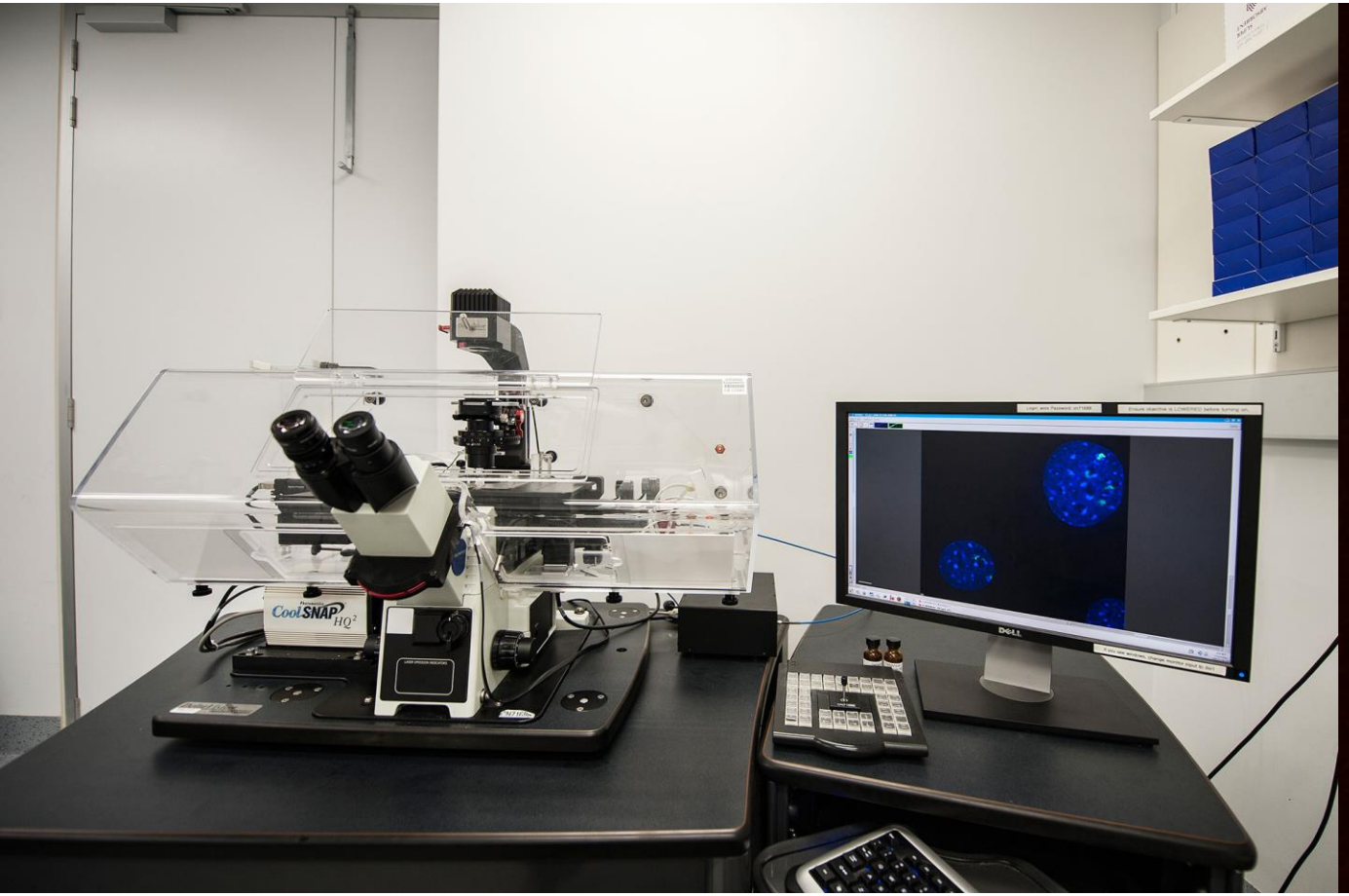
Figure 1. Mon2/Dop1 Complex 6Å Structure a) Mon2-TAP purification SDS-PAGE gel with input and elution fraction lanes after SEC purification. a-inset) zoomed in image of the elution fraction band displaying a doublet, indicative of the Mon2/Dop1 complex. **b)** unrefined 3D model from cryo-EM. **c)** refined 3D model from cryo-EM with corrected AF model of Mon2/Dop1 docked – view of Dop1. Structure corrected by having the two monomers have a trans-interaction at their N- and C-termini. Two different shades of pink depict different monomers of Dop1. **d)** same structure model as b, but 90˚turn to show Mon2. **e and f)** the same structure model as b and c except with the incorrect structure prediction of Dop1 as predicted by the AF program. AF predicted a cis-interaction between the N- and C-termini. This is depicted here as the different monomers are forming rings that interface at the center instead of an elongated figure eight interaction.

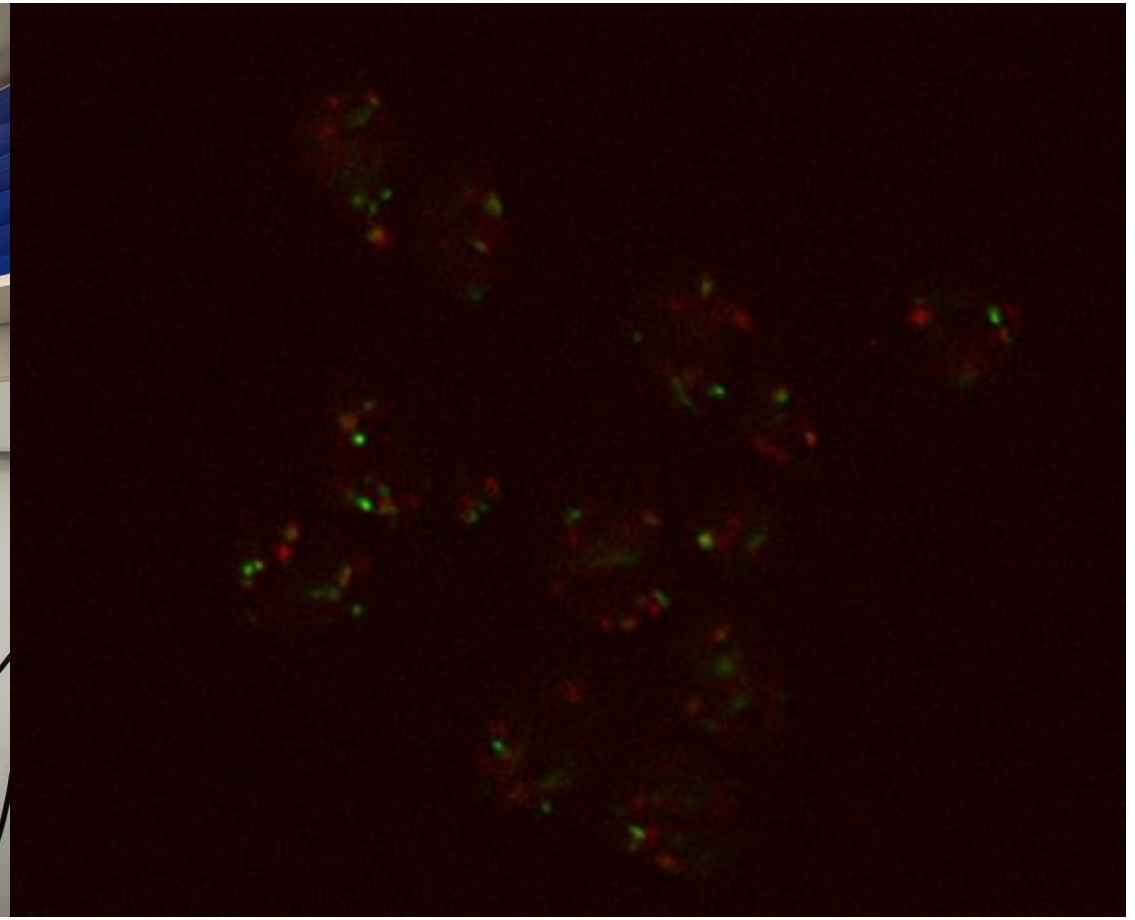
What do I spend my days doing? ***Cloning***



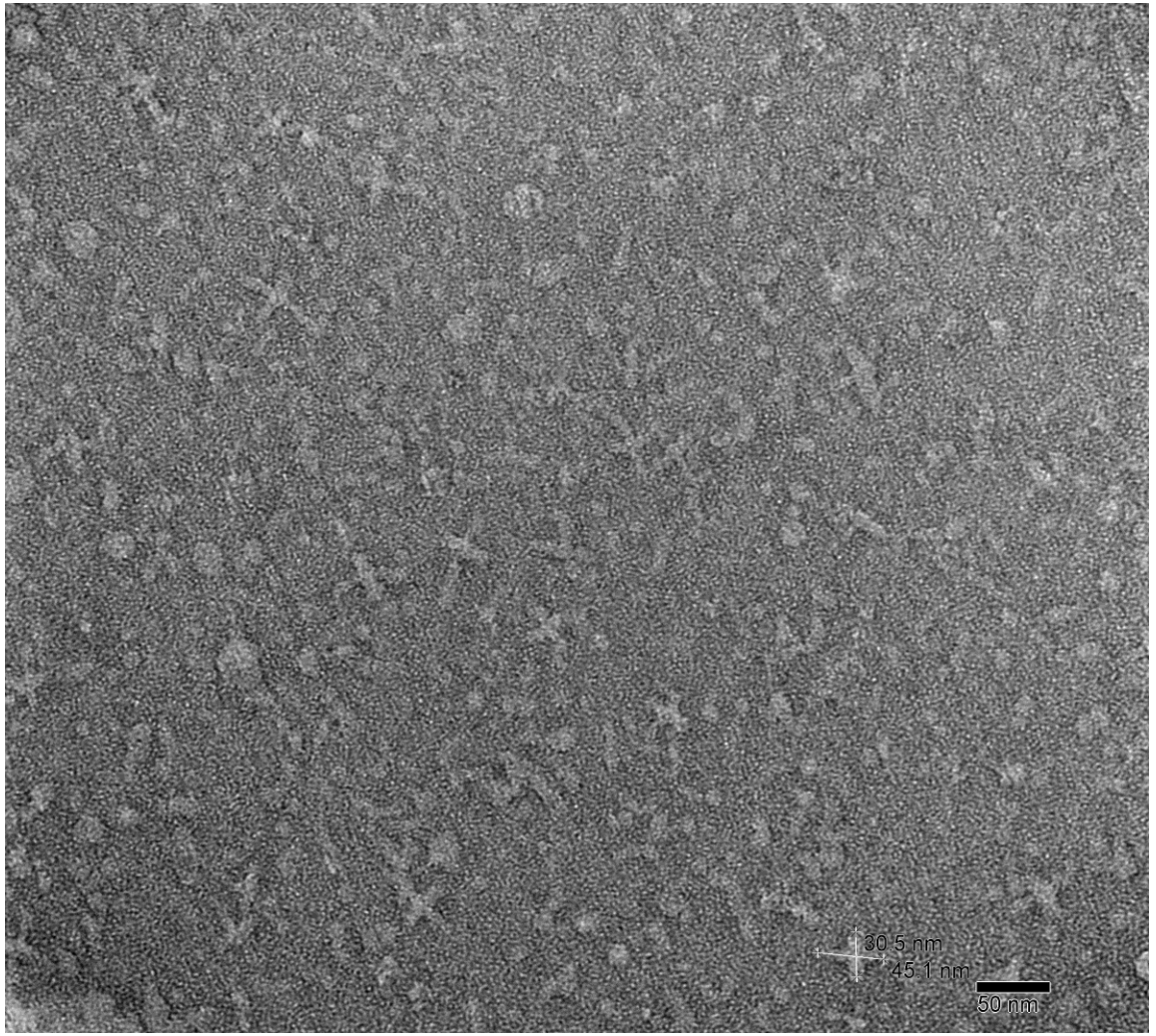


What do I spend my days doing? *Fluorescent Microscopy*





What do I spend my days doing? ***Cryo-EM***



What do I spend my days doing? *Protein Modelling*