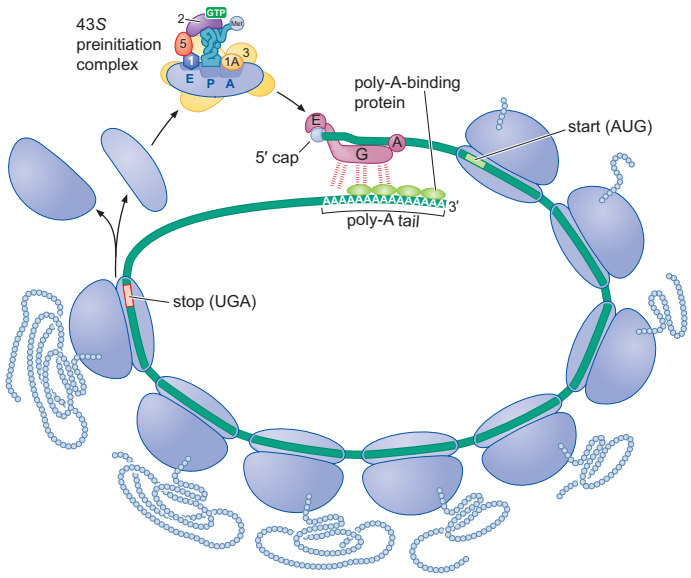
Lecture 11

Elongation and termination of prokaryotic cell

When the initiation complex forms, the appropriate aminoacyl-tRNA does not simply leave the synthetase and diffuse to the A site. Rather, it is delivered to the A site in association with a protein called elongation factor Tu named EF-Tu. This protein belongs to the G protein family. EF-Tu has two functions, first, protects the delicate ester linkage in aminoacyl-tRNA from hydrolysis. Second, EF-Tu is responsible for selection and binding of correct amino-acyl tRNA to the A site because GTP hydrolysis and expulsion of the EF-Tu-GDP complex from the ribosome occurs only if the pairing between the anticodon and the Condon is correct. Correct codon recognition induced an accommodation process which aligns the amino acids for peptide bond formation. With the formation of the peptide bond, the peptide chain is now attached to the tRNA whose anticodon is in the A site on the 30 subunit. However, protein synthesis cannot continue without the translocation of the mRNA AND the tRNAs within the ribosome. Elongation factor G which is named EF-G is responsible for the translocation of the ribosome one codon downstream. EF-G catalyzes the movement of mRNA, at the expense of GTP hydrolysis, by a distance of three nucleotides. So the peptidyl-tRNA moves out of the A site into the P site on the 30s subunits, and the deacylated tRNA moves from the P site into the E site and is subsequently released from the ribosome. Then the A site is exposed for the next aminoacyl-tRNA. The protein synthesis is terminated by release factors that read stop codons. There are three different release factors RF1,2 can pair with three different stop codons. RF3 is a GTPase that proofreads to ensure the stop codon is correctly recognised and stimulates the release of RF1 and 2

Translation of eukaryotic cell

Initiation: in eukaryotic, the initiation amino acid is methionine rather than N-formylmethionine. The aminoacyl-tRNA is called Met-tRNA$_i^{Met}$. The initiating codon in eukaryotes is always AUG which in the initiate sequence Kozak sequence. Initiation begins with the formation of a ternary complex consisting of the 40s ribosomes and Met-tRNA in association with eIF-2. The complex is called the 43s preinitiation complex (43 PIC), eLF-2 consists of **met-tRNA** and **2 GTP** molecules, and **binds to the vacant P site** in an unstable conformation, forming a **43S pre-initiation complex**



Initiation factor eIF-4E binds to the 5'-cap of the mRNA and facilitates binding of PIC to the mRNA. The association of eIF4G with eIF4E is particularly important—the overall level of translation in the cell is controlled at this step by a family of proteins that compete with eIF4G binding called eIF4E-binding proteins

This complex is joined by eIF4B, which activates the RNA helicase activity of eIF4A, eLF4A is a helicase activity, which cuts and separates any secondary structure on the mRNA to ensure smooth translation. The energy for the helicase activity is provided by **eIF4B**, which is a **GTPase**; hydrolysis of GTP into GDP and P_i. Soon after PIC binds the mRNA, eIF-4G links eIF-4E to a protein associated with the poly(A) tail, the poly(A)-binding protein I. Cap

and tail are thus brought together to form a circle of mRNA. The benefits of the circularisation of mRNA remain to be determined.

Elongation and termination: Eukaryotic elongation factors EF1alpha and EF1betagama are the counterparts of bacterial EF-Tu and EF-Ts. The GTP form of EF1alpha delivers aminoacyl-tRNA to the A site of the ribosome, and EF1betagama catalyzes the exchange of GTP for bound GDP. Eukaryotic EF2 mediates GTP-driven translocation in much the same way as bacterial EF-G. Termination in eukaryotes is carried out by a single release factor, eRF1, compared with two in bacteria, eRF1 can mimicking tRNA activity. it forces the ribosome to accept a water molecule, to cause a dissociation. Release factor eRF-3 accelerates the activity of eRF-1 by hydrolysis of GTP.