Unit 7: Translation (Prokaryotes and Eukaryotes)

Topic: Initiation in Eukaryotes

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Initiation in Eukaryotes

Initiation of translation in eukaryotes is similar to prokaryotic initiation in many ways:

- Both use a start codon and a dedicated initiator tRNA
- Both use initiation factors to form a complex with the small ribosomal subunit that assembles on the mRNA before addition of the large subunit.

Nevertheless, eukaryotes use a **fundamentally distinct method to recognize the mRNA and the start codon**, which has important consequences for eukaryotic translation.

The events of initiation can be broken down into four steps:

1. In contrast to the situation in prokaryotes, in eukaryotic cells, binding of the initiator tRNA to the small subunit always precedes association with the mRNA.

Refer to the 1st ribosome assembly figure of this slide, sent to you via group mail/Whatsapp.
2- A separate set of auxiliary factors mediates the recognition of the mRNA.

Refer to the initiation factor figure of this slide, sent to you via group mail/Whatsapp

3- The small ribosomal subunit bound to the initiator tRNA scans the mRNA for the first AUG sequence.

Refer to the figure showing difference between pro and eukaryotic ribosome binding and scanning, sent to you via group mail/Whatsapp

<table>
<thead>
<tr>
<th>In Prokaryotes</th>
<th>In Eukaryotes</th>
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<tr>
<td>RBS</td>
<td>AUG</td>
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4- The large subunit of the ribosome is recruited after the initiator tRNA base-pairs with the start codon.

Refer to the figure showing attachment of pre-initiation complex with that of larger subunit, sent to you via group mail/Whatsapp.
Refer to the FIGURE 15-26 Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA from Watson book - Molecular Biology of the Gene, sent to you via group mail/Whatsapp
eIF1, eIF1A, and eIF5 act in an analogous manner to the prokaryotic initiation factors IF3 and IF1 to prevent both large subunit binding and tRNA binding to the A-site.

eIF3 is almost as large as the entire 40S subunit but primarily binds the side of the small subunit near the RNA entry and exit sites. Nevertheless, eIF3 interacts with every member of the 43S PIC including the initiator tRNA and, thus, facilitates many of the interactions involved in 43S PIC assembly.

Refer to the FIGURE 15-26 Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA from Watson book- Molecular Biology of the Gene, sent to you via group mail/Whatsapp.
The initiator tRNA (here it is met, not formyl met) is escorted to the small subunit by the three subunit GTP-binding protein eIF2.

Like IF2, eIF2 will bind the initiator tRNA only in the GTP-bound state.

The complex between the initiator tRNA and EIF2 is called the ternary complex (TC).

eIF2 positions the Met-tRNAi Met in the P-site of the initiation factor–bound small subunit, resulting in the formation of the 43S preinitiation complex (43S PIC)

Refer to the FIGURE E 15-26 a Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA from Watson book- Molecular Biology of the Gene, sent to you via group mail/Whatsapp
In a separate series of reactions, the mRNA is prepared for recognition by the small subunit recognition of the 5’ cap by the cap-binding protein eIF4E.

eIF4G binds to both eIF4E and the mRNA, whereas eIF4A binds eIF4G and the mRNA

eIF4B, activates the RNA helicase activity of eIF4A. Helicase unwinds any secondary structures (such as hairpins) that may have formed at the end of the mRNA.

Refer to the FIGURE 15-26 b Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA from Watson book- Molecular Biology of the Gene, sent to you via group mail/Whatsapp.
Finally, interactions between the eIF4G bound to the unstructured mRNA and the initiation factors (particularly eIF3) bound to the small subunit recruit the 43S preinitiation complex to the mRNA to form the 48S preinitiation complex.

Translation regulation step- at this step by a family of proteins that compete with eIF4G binding called eIF4E-binding proteins

Refer to the FIGURE 15-26 Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA from Watson book- Molecular Biology of the Gene, sent to you via group mail/Whatsapp
Translation Initiation Factors Hold Eukaryotic mRNAs in Circles

The presence of a poly-A tail contributes to the efficiency of eukaryotic Translation.

eIF4G binds both directly to the 3’ end of the mRNA and to the poly-A-binding protein that coats the poly-A tail.

Benefits:

- Large subunit recruitment.
- mRNA are maintained through multiple rounds of translation.
- Added benefit of locating recently terminated ribosomes near the AUG, presumably enhancing reinitiation.

Refer to FIGURE 15-27 “A model for the circularization of eukaryotic mRNA. Circularization is mediated by interactions between eIF4G, the poly-A-binding protein, and the poly-A tail.” from Watson book Molecular Biology of the Gene, sent to you via group mail/Whatsapp.
Let’s move towards Translation Elongation !!!