

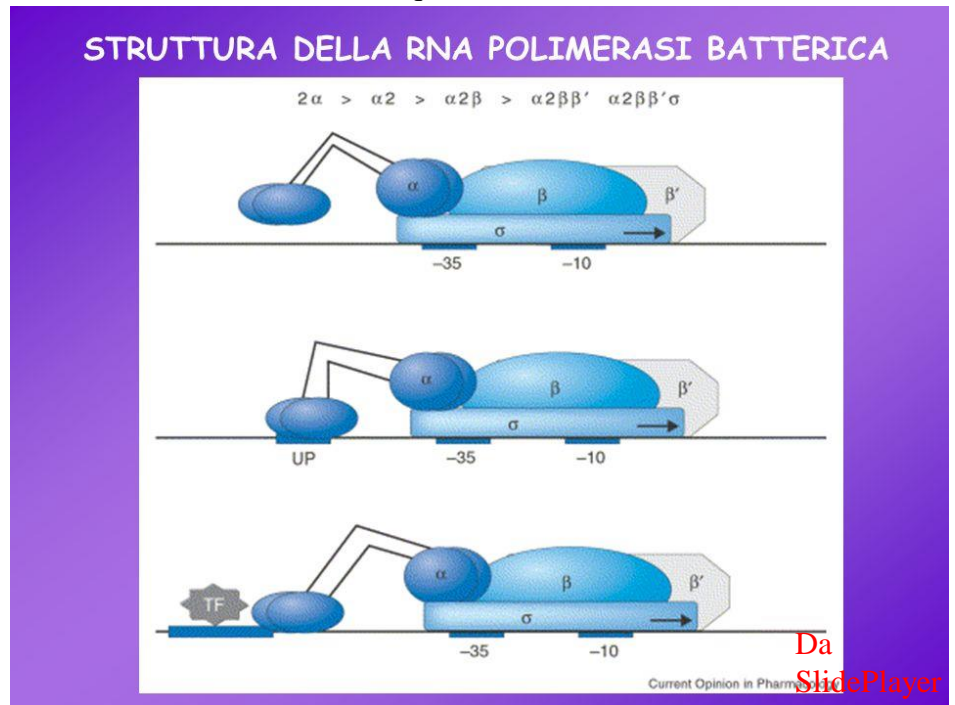
# SINTESI PROTEICA

TUTORATO 11 | GIULIANO F. PATANÈ | COLLEGIO A. VOLTA

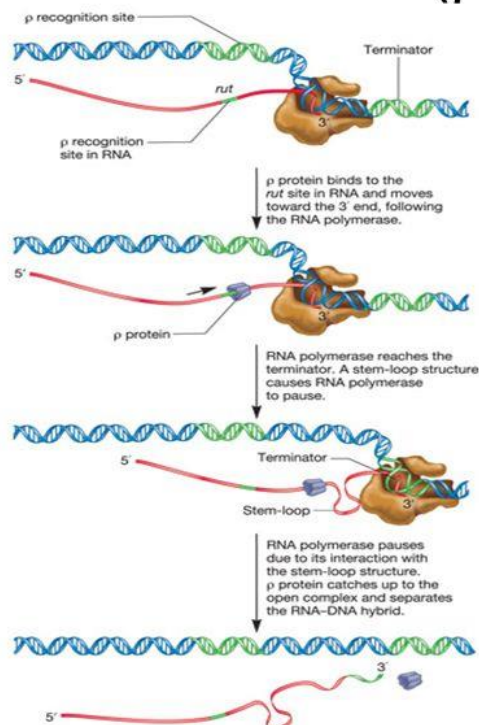
# Rna polimerasi

L'enzima RNA polimerasi batterica è costituito da cinque subunità:

- 2 subunità beta;
- 1 subunità omega;
- 2 subunità alfa.



## Terminazione dipendente dal fattore Rho ( $\rho$ )



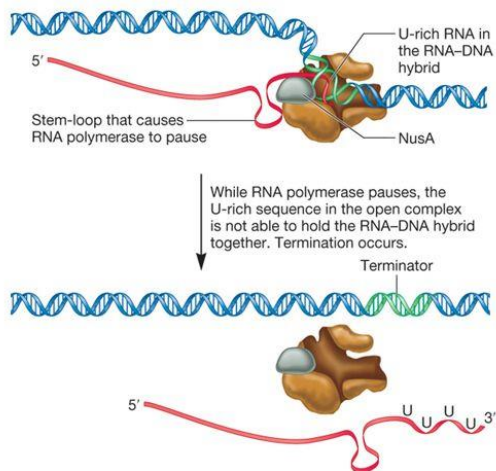
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# Terminazione rho indipendente

La terminazione della trascrizione avviene secondo due diverse modalità:

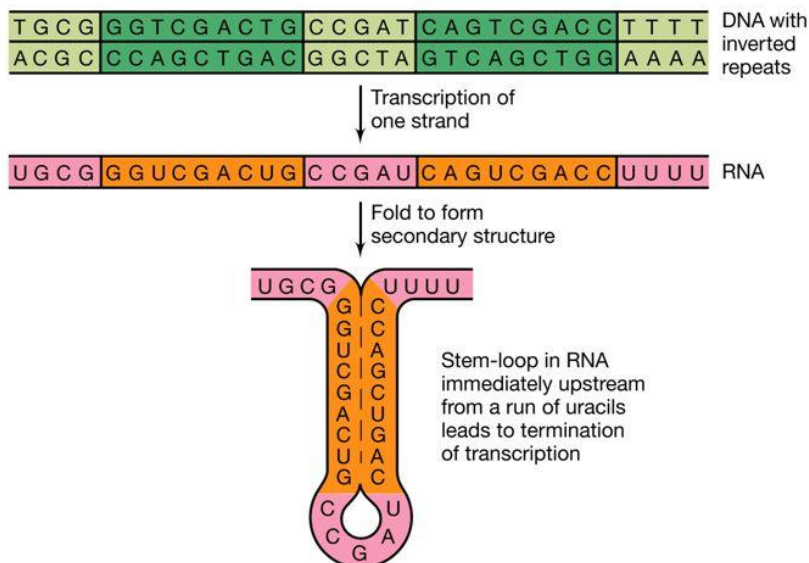
- $\rho$  dipendente;
- $\rho$  indipendente.

## Terminazione "intrinseca" (rho-indipendente)



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## Come si ferma una macchina in corsa? Il terminatore

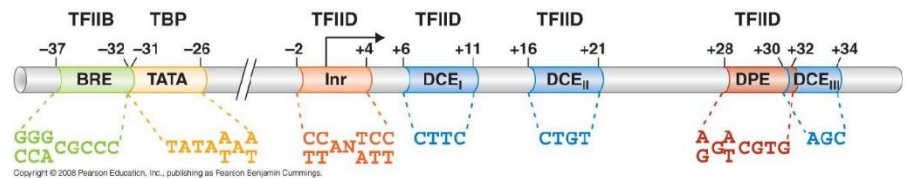


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# Trascrizione eucarioti

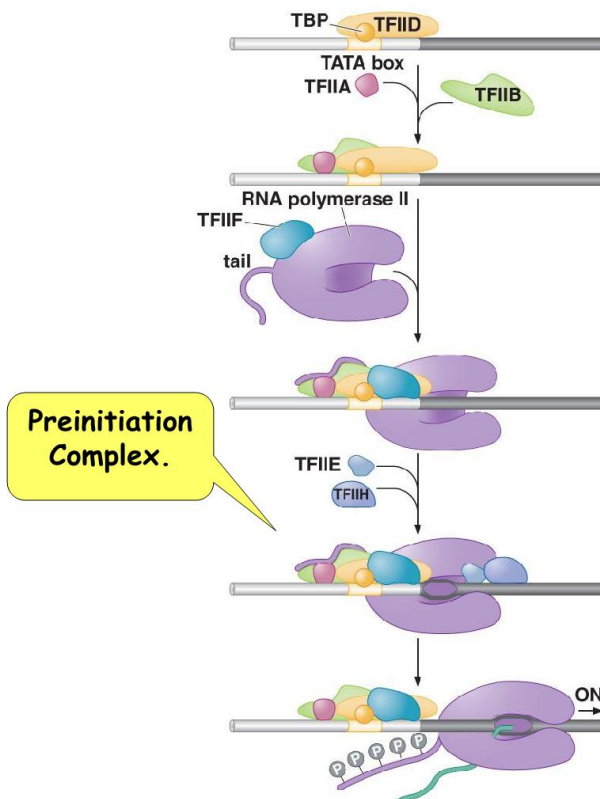
L'inizio della trascrizione negli eucarioti è regolata da una serie di proteine definite fattori di trascrizione (FTII)

Transcription in Eukaryotes needs several initiation factors called general transcription factors (GTFs)



CORE PROMOTER

## Transcription initiation by RNA Pol II



The opening of the Preinitiation Complex in eukaryotes requires ATP hydrolysis for DNA melting and it is due to TFIIH factor. The RNA escaped from the promoter also requires energy and phosphorylation of the polymerase tail (CTD: carboxy terminal domain).

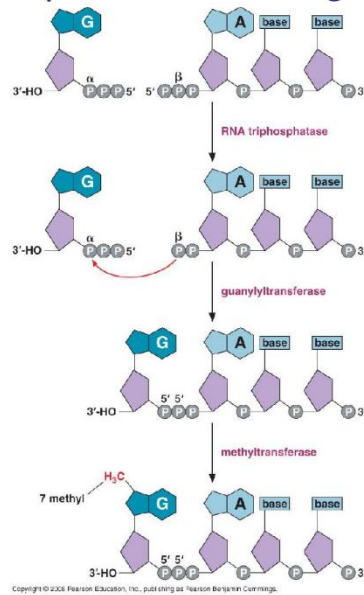
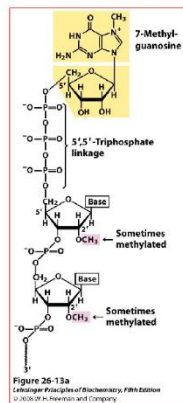
# MATURAZIONE TRASCritto

Il processamento dell'RNA messaggero negli eucarioti consta di tre tappe:

- capping;
- poliadenilazione;
- splicing.

**5'-end RNA Capping: Necessary to protect RNA from rapid degradation and to help ribosomes binding.**

It happens as soon as the RNA emerges from the RNA-exit channel.

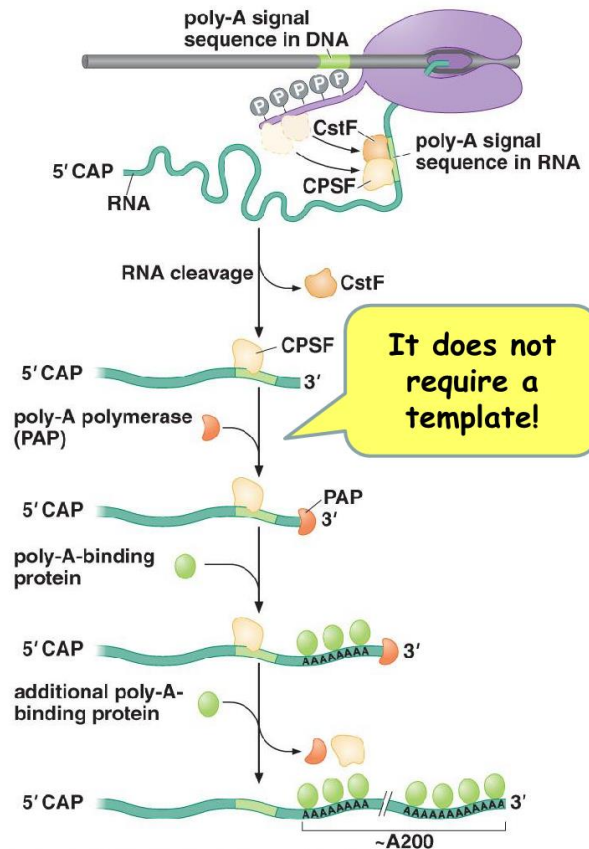


## 3'-end RNA polyadenylation

**CPSF:** Cleavage and polyadenylation Specificity factor

**CstF:** cleavage stimulator factor

It protects RNA from enzymatic degradation!



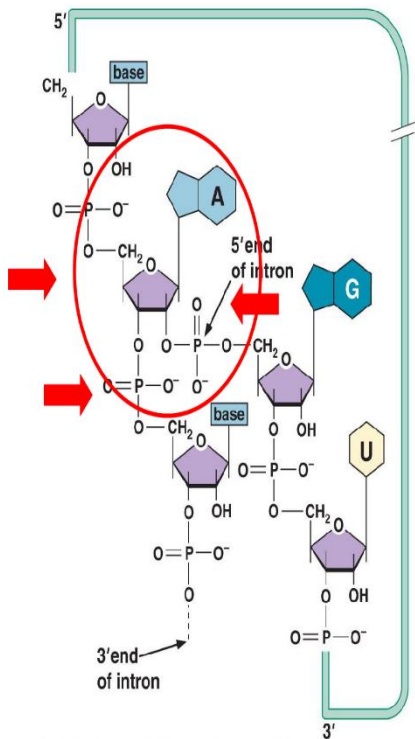
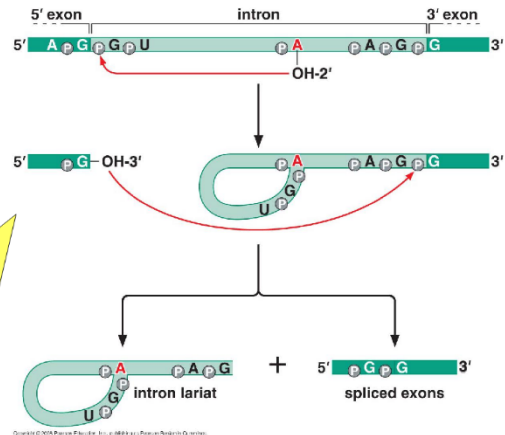
# splicing

Il processo di **splicing** è permesso grazie alla presenza di tre sequenze altamente conservate negli introni dell'mRNA:

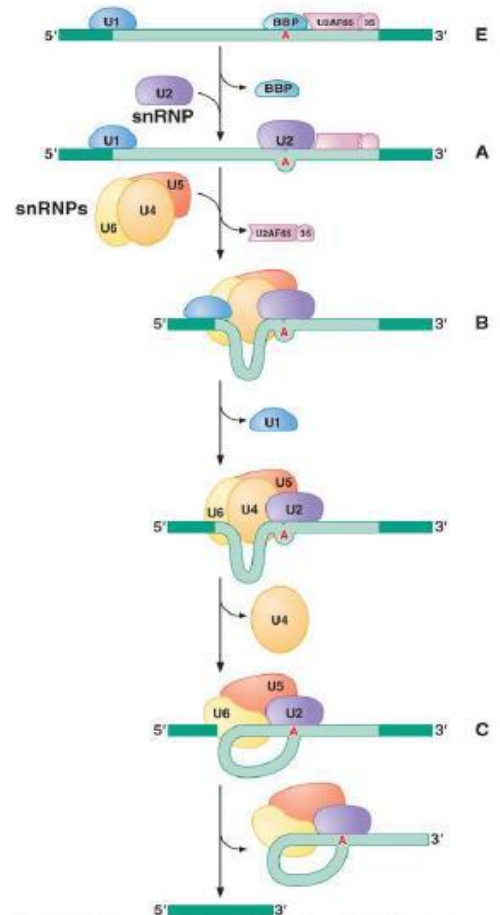
- GU al 5';
- AG al 3';
- A (adenina del branch site) a circa 20 nucleotidi dal 3'.

## The Chemistry of Splicing

The splicing requires two successive transesterification! Two chemical bonds are broken and two new ones are formed. No energy is required for these reactions! Indeed splicing process itself requires high amount of energy for the spliceosome assembly and proper activity.



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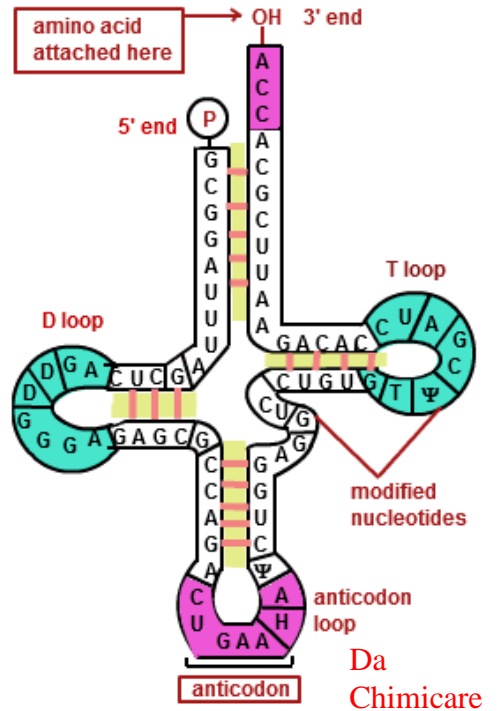


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# Rna transfer

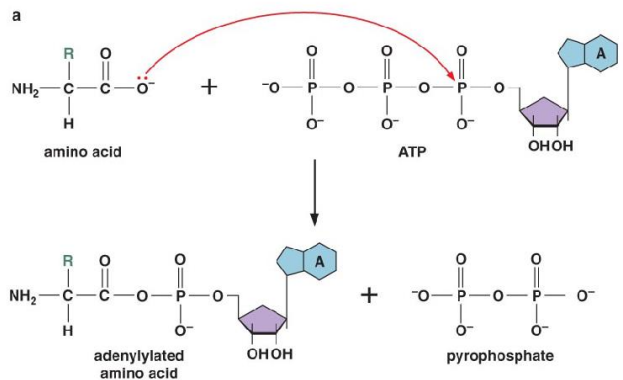
La struttura secondaria a trifoglio del t-RNA è caratterizzata da quattro bracci:

- braccio accettore;
- braccio D;
- braccio A;
- braccio T.

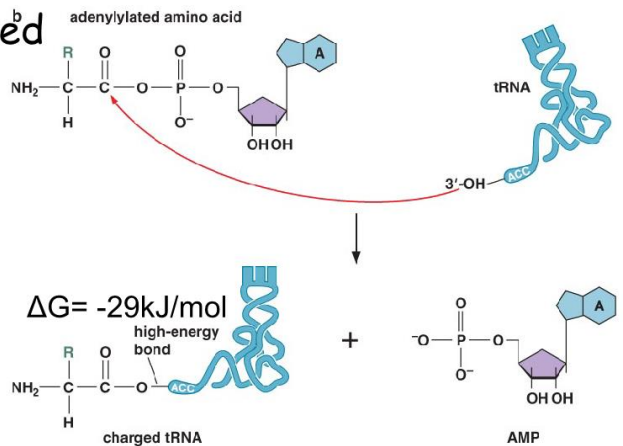
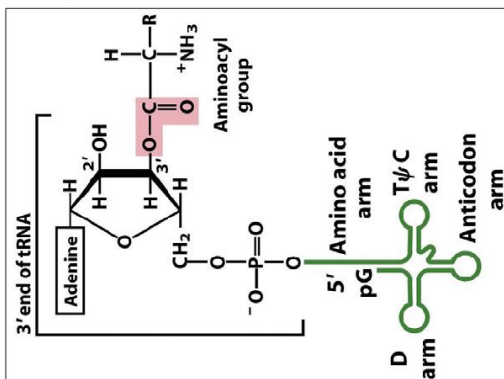


Aminoacyl-tRNA charging:

1) adenylation



2) transfer of the adenylylated amino acid to tRNA



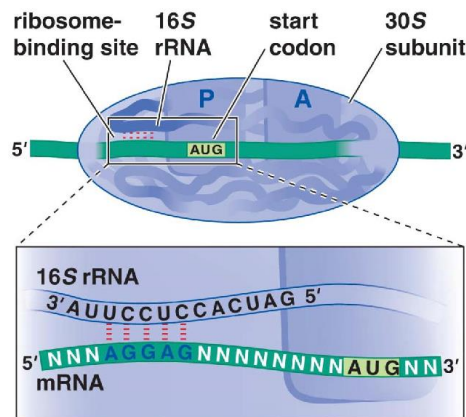
# RIBOSOMA

Il **ribosoma** presenta tre differenti siti di legame per i t-RNA:

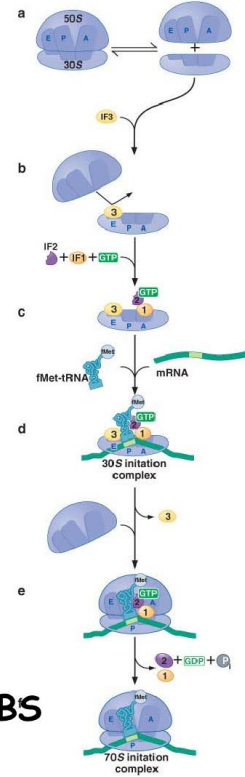
- sito E;
- sito P;
- sito A.

## Translation initiation in prokaryotes

- Ribosome recruitment to the mRNA;
- a charged tRNA (fMet-tRNA) is placed in the P site;
- the ribosome is positioned over the start codon

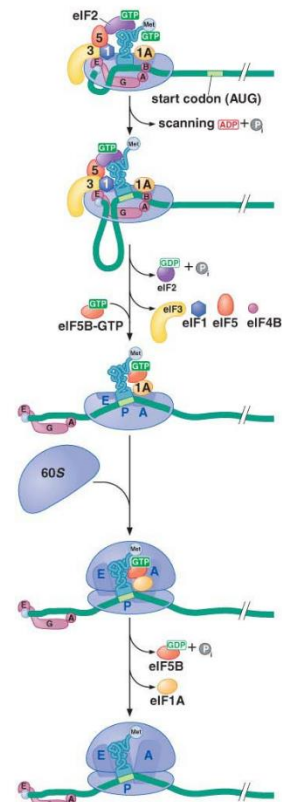
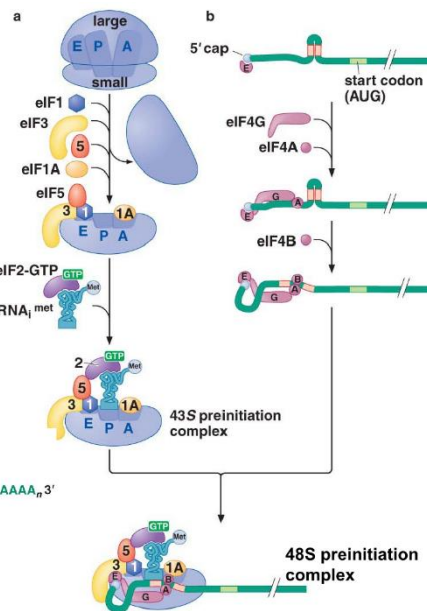
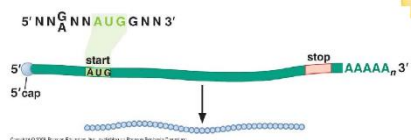


The 16S rRNA interacts with the RBS to position the AUG in the P site.



## Translation initiation in eukaryotes

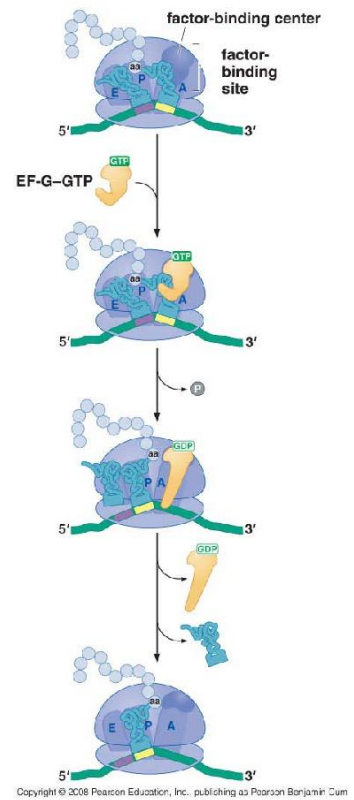
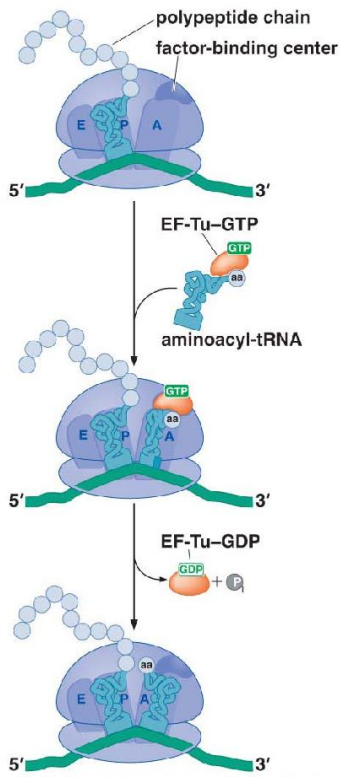
Assembly of the eukaryotic small ribosomal subunit and initiator tRNA onto the mRNA





# Allungamento e terminazione

Lo spostamento del ribosoma lungo l'm-RNA avviene grazie al fattore EF-G (negli eucarioti, EF2).



# TERMINAZIONE

La **terminazione** della traduzione prevede l'azione dei fattori RF-1 e RF-3. Affinché, invece, il ribosoma si dissocia nelle sue due subunità costituenti, entra in gioco RRF (ribosom relasing factor) e IF-G.

