TRASCRIZIONE NEGLI EUCARIOTI

CLASSES OF EUKARYOTIC CELLULAR RNAS

- ribosomal RNA (rRNA)
  - 18S (small subunit)
  - 28S (large subunit)
  - 5.8S (large subunit)
  - 5S (large subunit)
- transfer RNA (tRNA)
- messenger RNA (mRNA)
- heterogeneous nuclear RNA (hnRNA) (precursors of mRNA)
- small nuclear RNA (snRNA)
  - U1, U2, U3, U4, U5, U6, U7, U8, U9, U10...
- small nucleolar RNA (snoRNA)
- small cytoplasmic RNA (scRNA)
  - 7SL RNA
- miRNA
- long non-coding RNA (lncRNA)

THE HUMAN RNA POLYMERASES

Polymerase	Location	Product
RNA polymerase I	nucleolus	18S, 28S, 5.8S rRNA
RNA polymerase II	nucleoplasm	hnRNA/mRNA, U1, U2, U4, U5 snRNA, miRNA, lncRNA
RNA polymerase III	nucleoplasm	tRNA, 5S RNA, U6 snRNA, 7SL, miRNA
mitochondrial RNA polymerase	mitochondrion	all mitochondrial RNA

RNA POLIMERASI EUCARIOTICHE

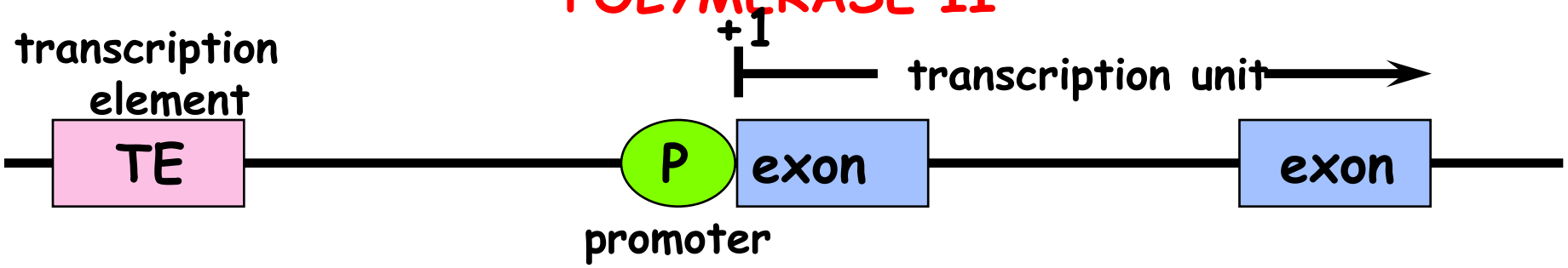
Enzimi formati da numerose subunita'.

Differiscono anche per la sensibilita' all' a-amanitina
RNA pol I resistente
RNA pol II alta sensibilita' (Kd = 10^{-8} M)
RNA pol III bassa sensibilita' (Kd = 10^{-6} M)

Polimerasi II: il dominio carbossiterminale (CTD) della subunita' maggiore ha numerose ripetizioni di una sequenza di 7 aa (di cui 3 serine).

Riconoscono promotori diversi.

TRANSCRIPTION AND PROMOTER ELEMENTS FOR RNA POLYMERASE II



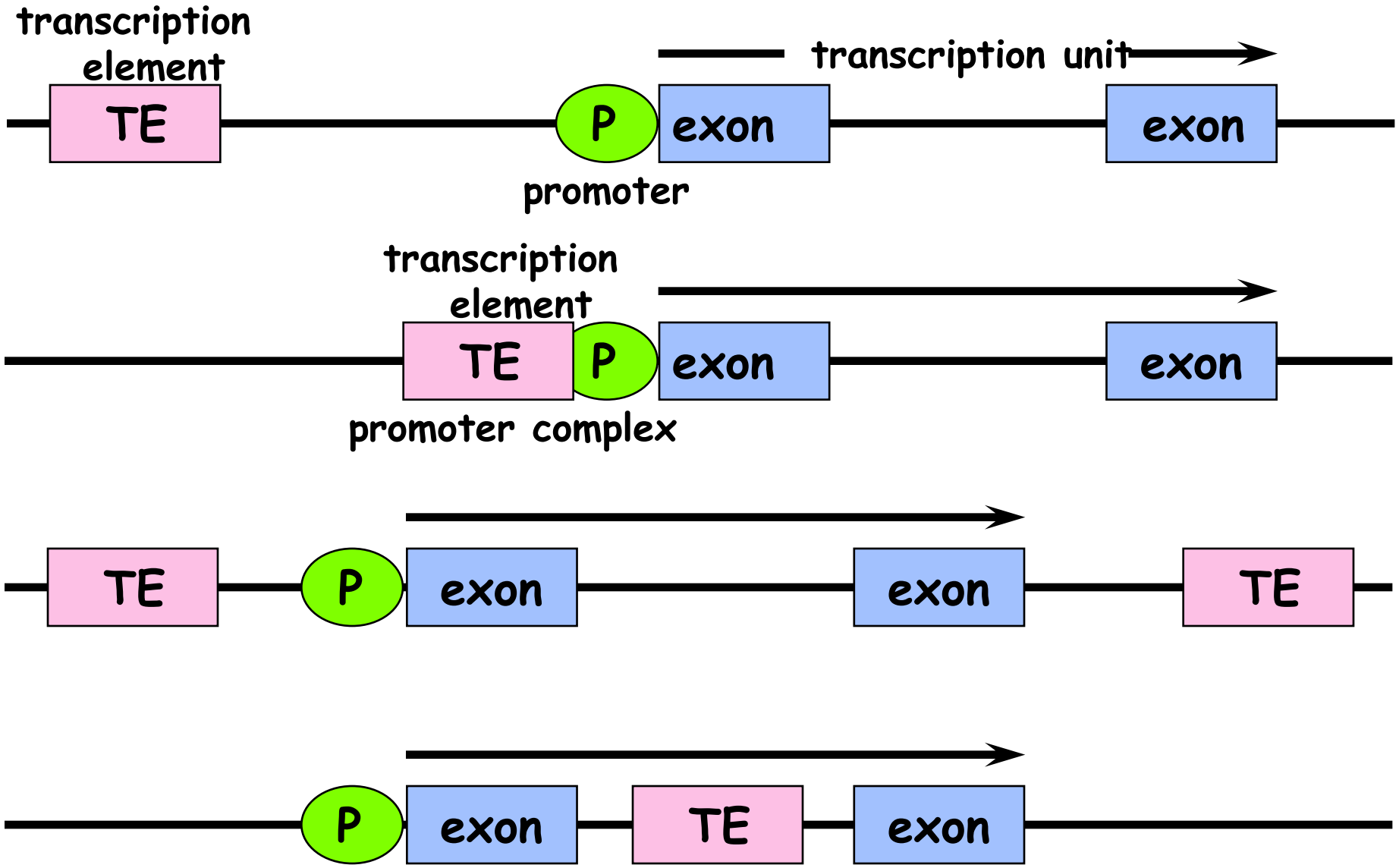
Promoter (DNA sequence upstream of a gene)

- determines start site (+1) for transcription initiation
- located immediately upstream of the start site
- allows basal (low level) transcription

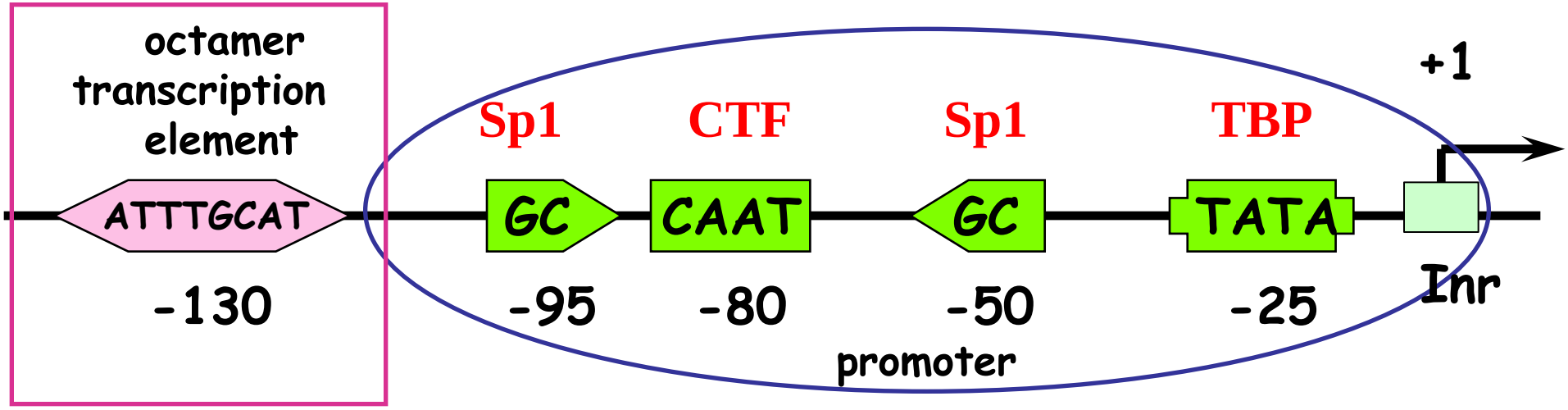
Transcription element (DNA sequence that regulates the gene)

- determines frequency or efficiency of transcription
- located upstream, downstream, or within genes
- can be very close to or thousands of base pairs from a gene
- includes
  - enhancers (increase transcription rate)
  - silencers (decrease transcription rate)
  - response elements (target sequences for signaling molecules)
- genes can have numerous transcription elements

TRANSCRIPTION AND PROMOTER ELEMENTS FOR RNA POLYMERASE II



SEQUENCE ELEMENTS WITHIN A TYPICAL EUCARYOTIC GENE PROMOTER



TATA box (TATAAAA)

- located approximately 25-30 bp upstream of the +1 start site
- determines the exact start site (not in all promoters)
- binds the TATA binding protein (TBP) which is a subunit of TFIID

GC box (CCGCCC)

- binds Sp1 (Specificity factor 1)

CAAT box (GGCCAATCT)

- binds CTF (CAAT box transcription factor)

Octamer (ATTTGCAT)

- binds OTF (Octamer transcription factor)

PROTEINE NECESSARIE PER L'ATTIVITA' DELLA POLIMERASI II

Fattori di trascrizione generali (TFII): necessari per qualsiasi promotore della Pol II.

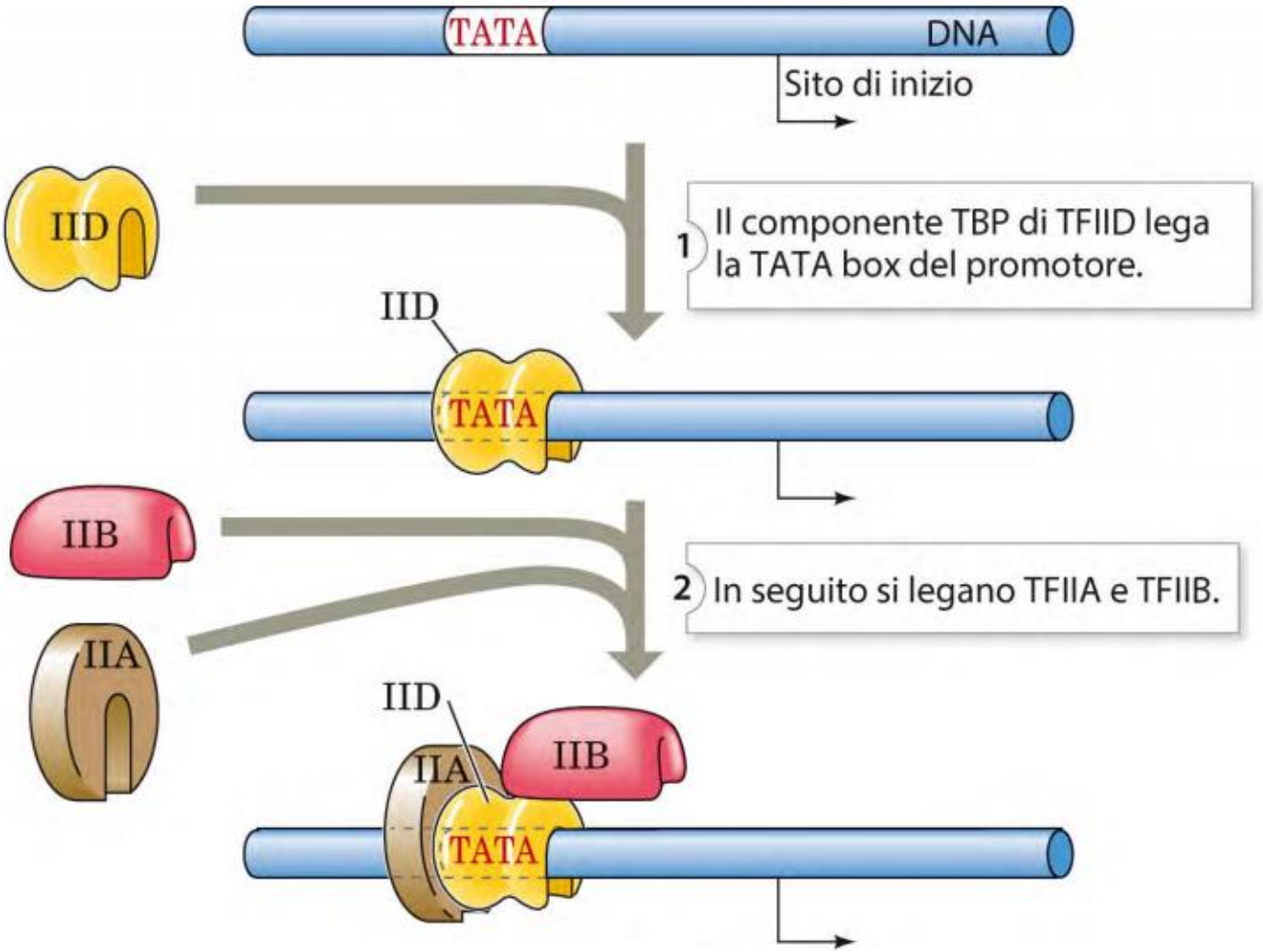
- Fattori di inizio:
• TFIID (che comprende diverse subunita' tra cui la TBP, che lega la TATA box)
  •TFIIA, TFIIB, TFIIE, TFIIF, TFIIH[1]

intervengono nella fase di inizio: formazione del complesso di inizio della trascrizione a livello delle sequenze promotrici.

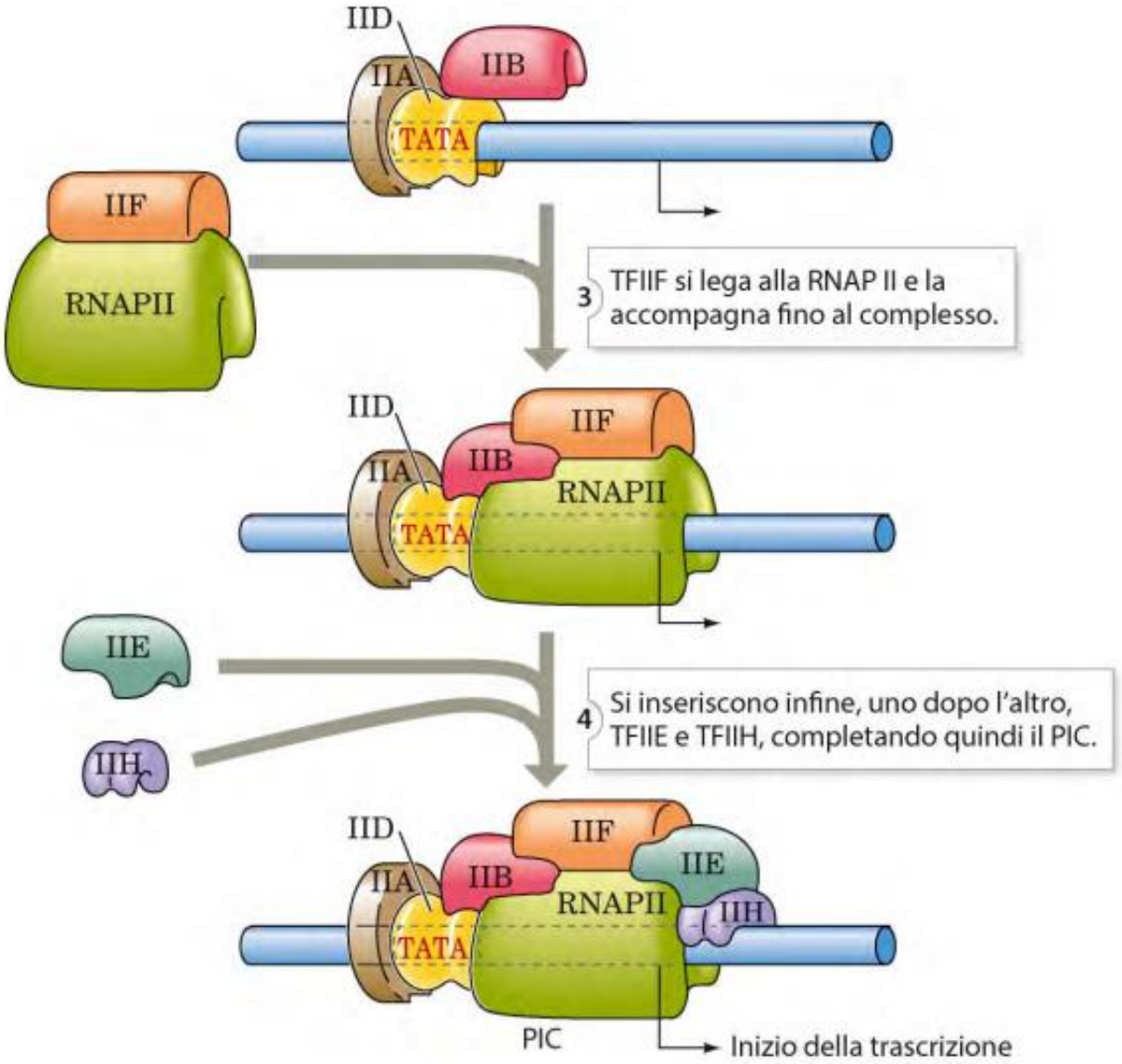
-Fattori di allungamento
-Fattori di termine

[1]TFIIH ha attivita' **elicasica**, e' coinvolto nella **fosforilazione** della Pol II e nella **riparazione del DNA** (excisione di nucleotidi).

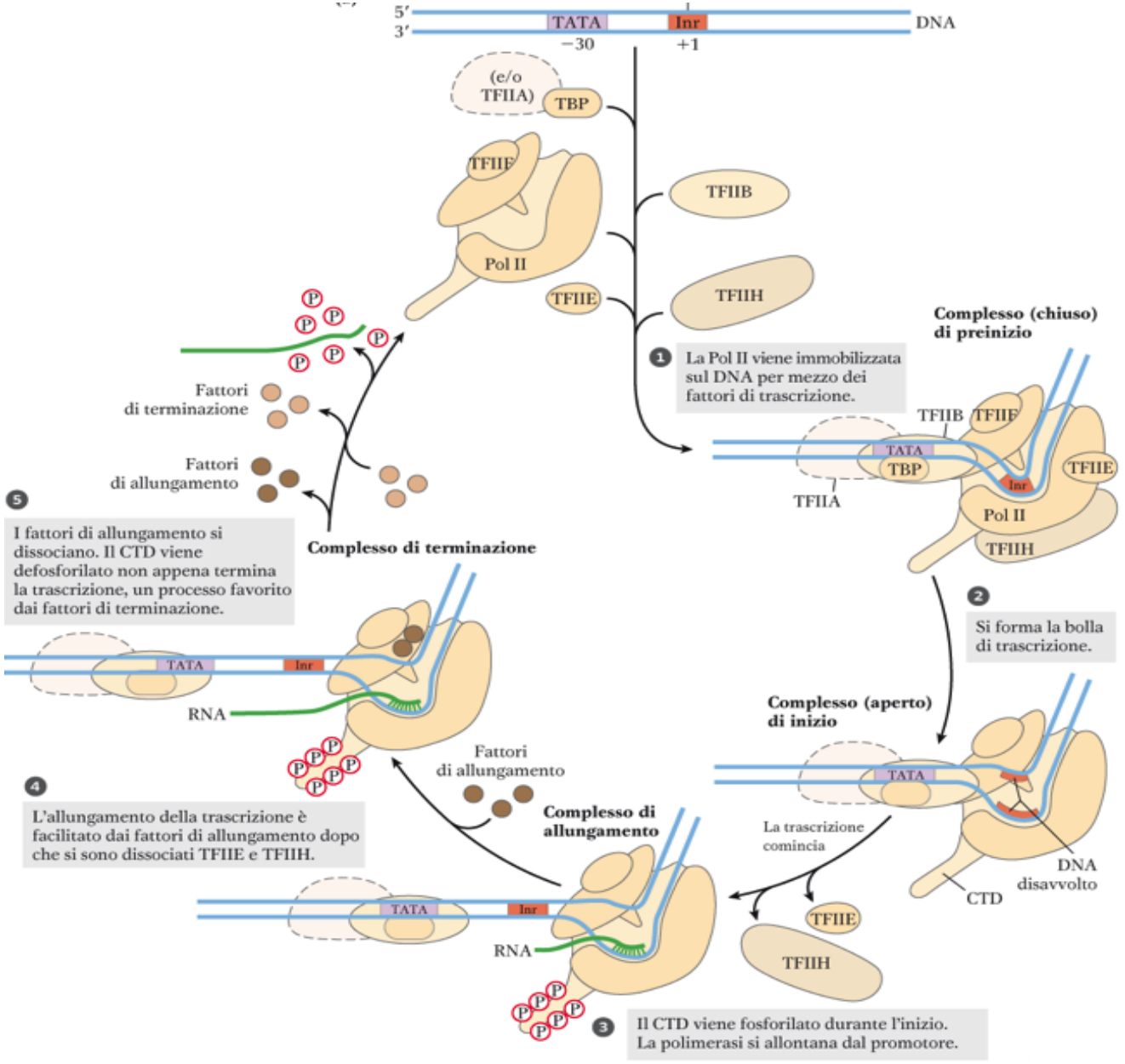
Formazione del complesso di preinizio della trascrizione promotore RNA polimerasi II (1)



Formazione del complesso di preinizio della trascrizione promotore RNA polimerasi II (2)



Trascrizione a livello dei promotori della RNA polimerasi II

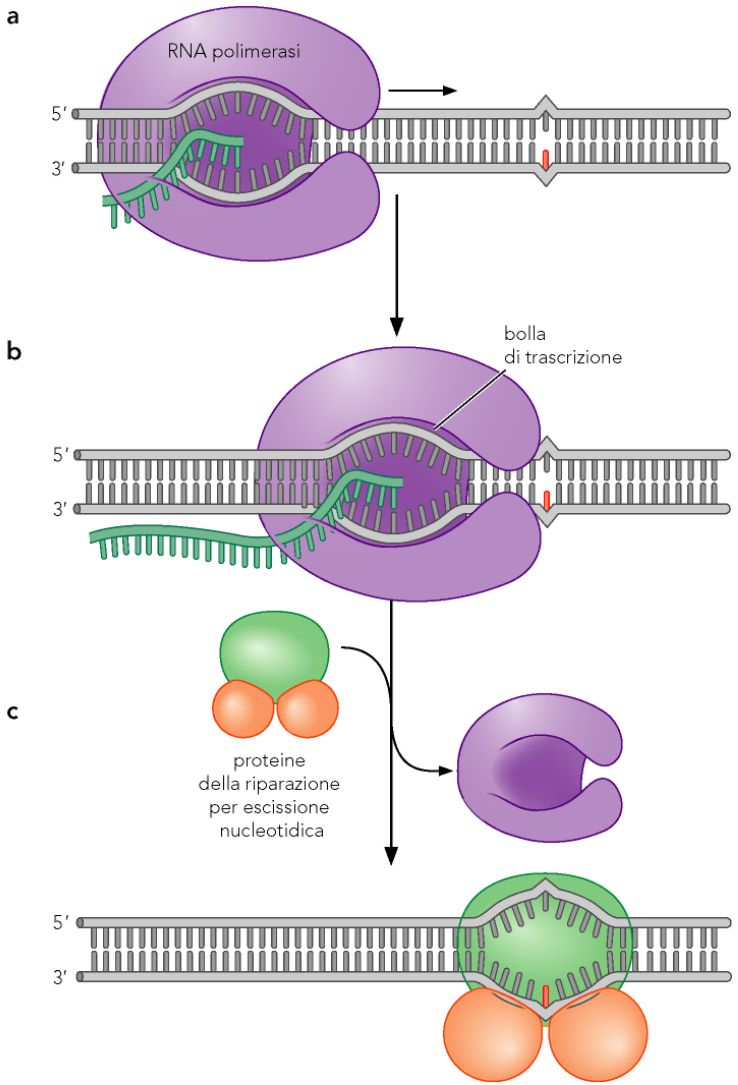


Nelson & Cox I principi di Biochimica di Lehninger- Zanichelli 6 ed.

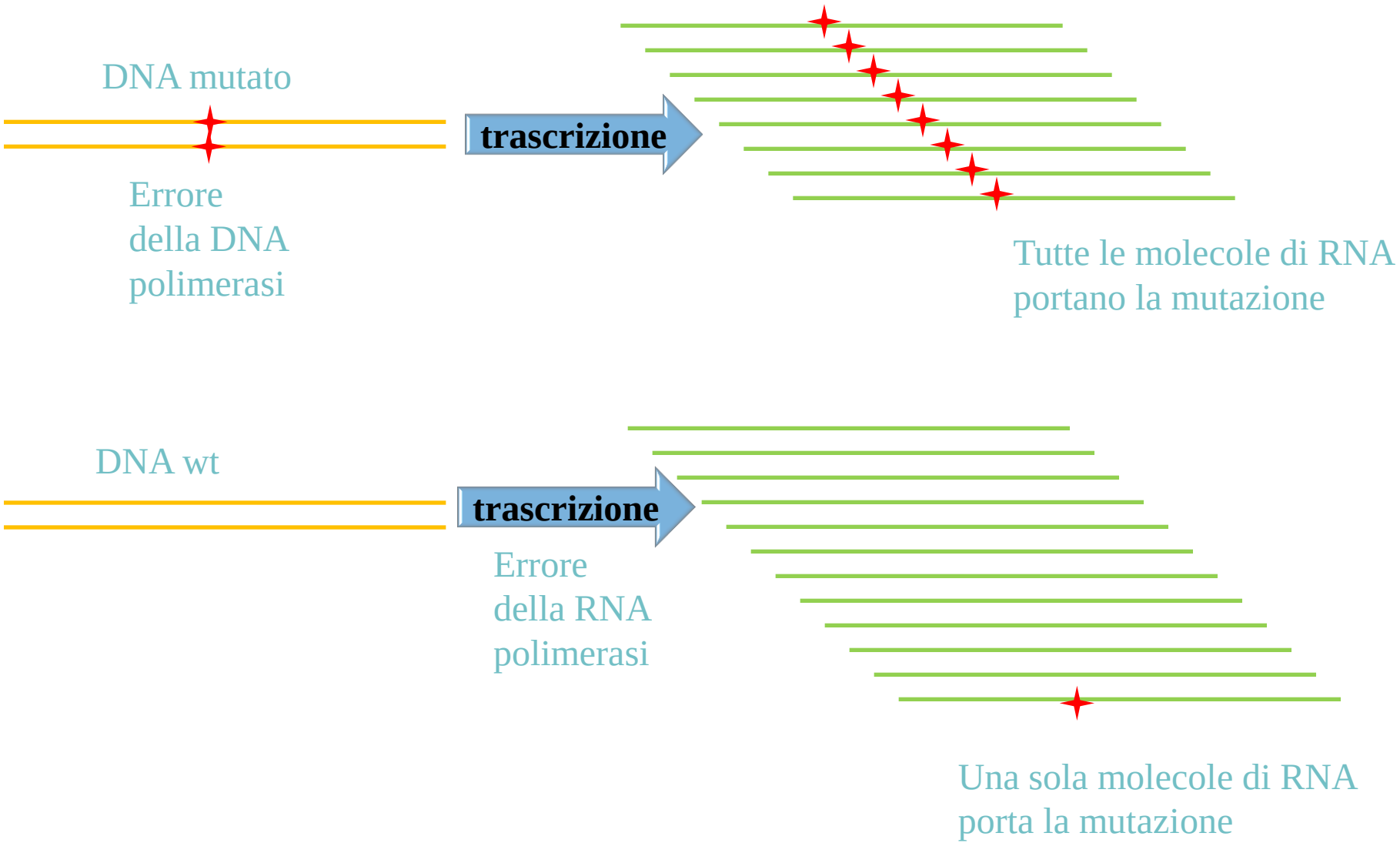
Transcription-Coupled NER (nucleotide excision repair)

Negli eucarioti la riparazione del DNA danneggiato è piu' efficiente per i geni che vengono attivamente trascritti.

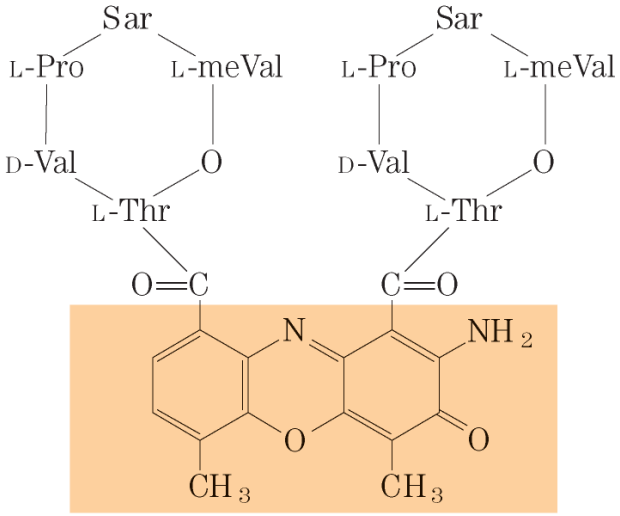
Ruoli del fattore TFIIH



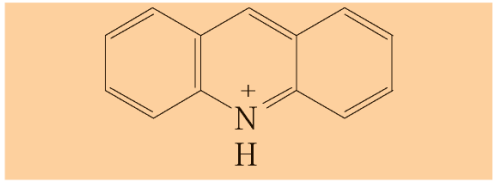
Effetti di errori delle polimerasi



Inibitori della trascrizione sia in Procarioti che Eucarioti

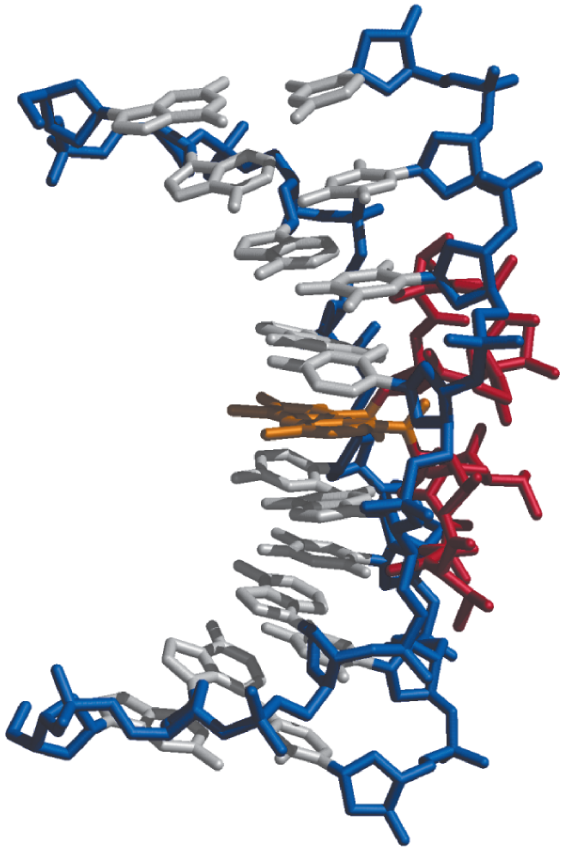


Actinomicina D



Acridina

(a)



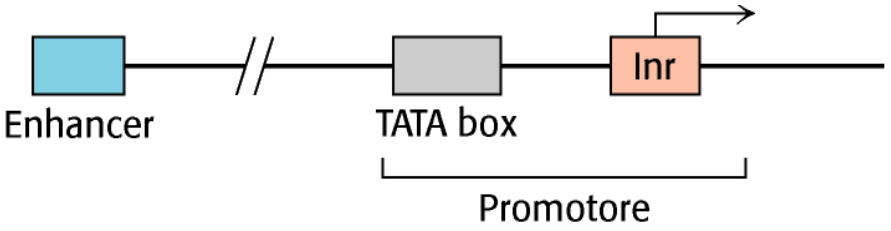
(b)

Sia l'actinomicina D che l'acridina sono intercalanti, che bloccano l'allungamento della catena da parte della RNA polimerasi

Nelson & Cox I principi di Biochimica di Lehninger- Zanichelli 6 ed.

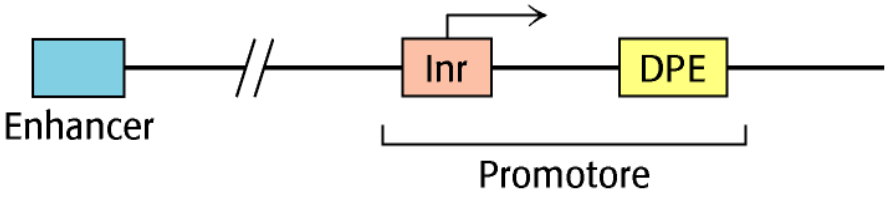
Promotori RNA polimerasi eucariotiche

RNA PolII



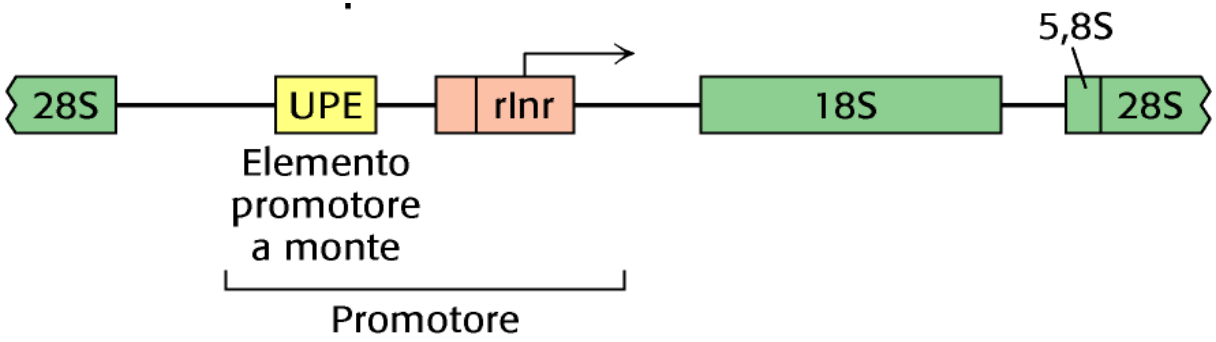
Inr= initiator element

oppure

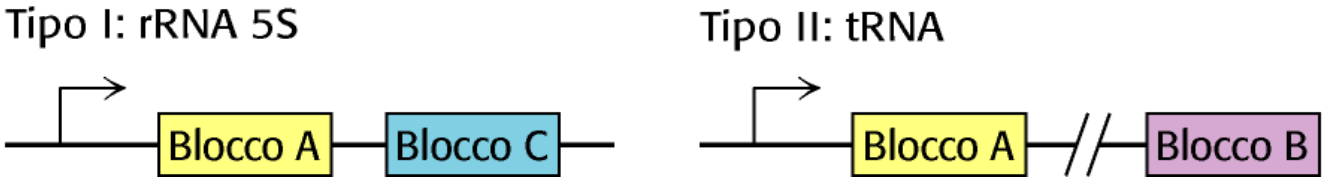


DPE= downstream promoter element

RNA PolI



RNA PolIII



Berg J, Tymoczko JL, Stryer L. Biochimica- Zanichelli 6 ed