EUKARYOTIC TRANSCRIPTION FACTORS
(trans-acting factors)

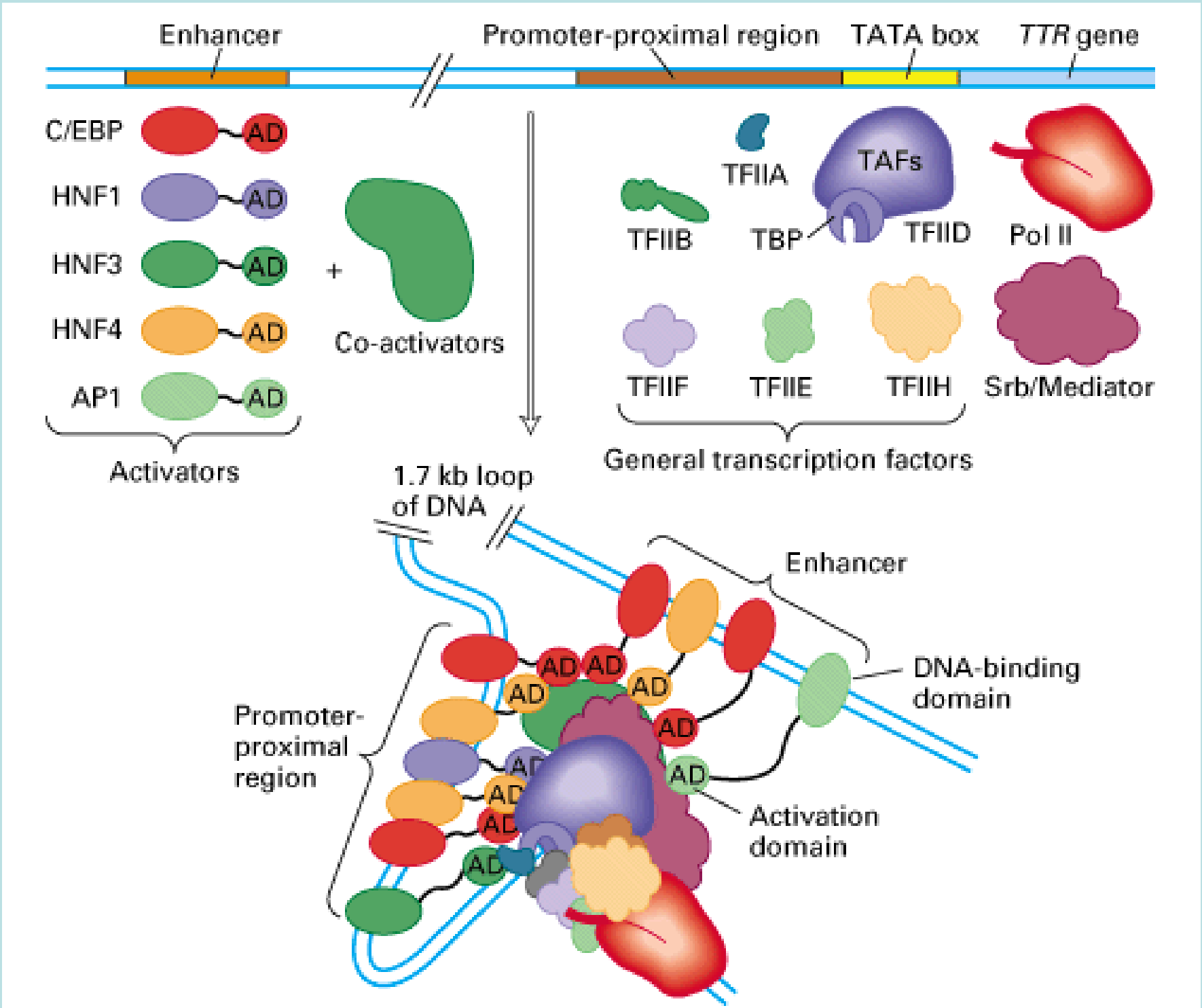
Can be divided in:

- General transcription factors

- Activators/inactivators

Activators

- They can be constitutively expressed o induced.
- They can be ubiquitary expressed or tissue-specific.
- They recognize specific DNA sequences named Response Elements (REs) that can be found in the promoter or enhancer of more than one gene.
- They can act interacting with coattivators.


Enhancer
Promoter-proximal region
TATA box
TTR gene
C/EBP
HNF1
HNF3
HNF4
AP1
AD
Co-activators
Activators
TFIIA
TAFs
TFIIB
TBP
TFIID
Pol II
TFIIF
TFIIE
TFIIH
Srb/Mediator
General transcription factors
1.7 kb loop of DNA
Enhancer
DNA-binding domain
Promoter-proximal region
Activation domain

The induced transcription factors can be bound to: response elements identifying groups of promoters, or enhancer subjected to a coordinate control

Regulatory agent	Module	Consensus sequence	Factor	Weight (dalton)
Heat shock	HSE	CNNGCCNNTCCNNG	HSTF	93,000
Glucocorticoid	GRE	TGGTACAAATGTTCT	Recettore	94,000
Cadmium	MRE	CGNCCCGGNCNC	?	?
TPA	TRE	TGACTCA	AP1	39,000
Serum	SRE	CCATATTAGG	SRF	52,000

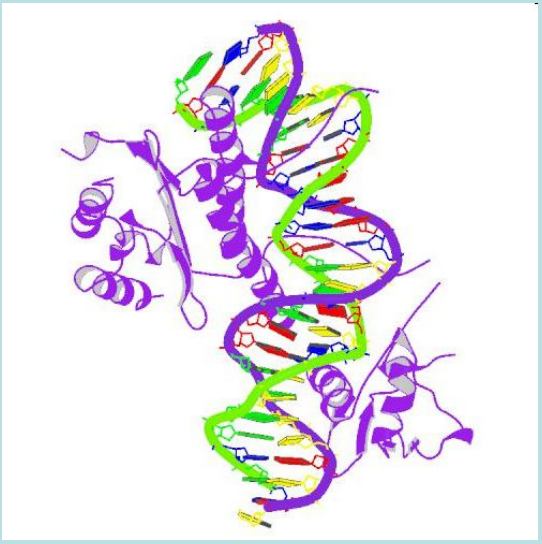
Heat Shock Response Element

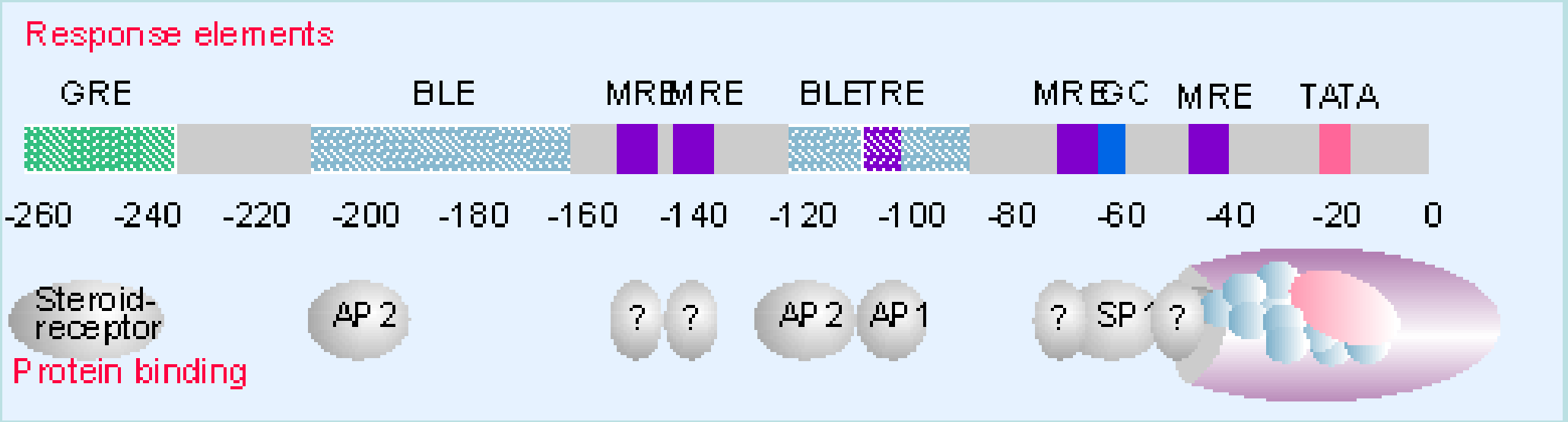
Glucocorticoid Response Element

Metal Response Element

12-O-Tetradecanoylphorbol-13-acetate Response Element

Serum Response Element





The regulatory region of a human metallothionein gene contains regulator elements in both its promoter and enhancer. The promoter has elements for metal induction; an enhancer has an element for response to glucocorticoid. Promoter elements are shown above the map, and proteins that bind them are indicated below.

GRE => Glucocorticoid Response Element

BLE => Basal Level Element

MRE => Metal Response Element

TRE => TPA Response Element

TRANSCRIPTION FACTORS HAVE A "MODULAR" STRUCTURE MADE BY DIFFERENT DOMAINS

- **Binding to DNA**
- **Transactivation**
- **Dimerization**
- **Binding to the ligand**

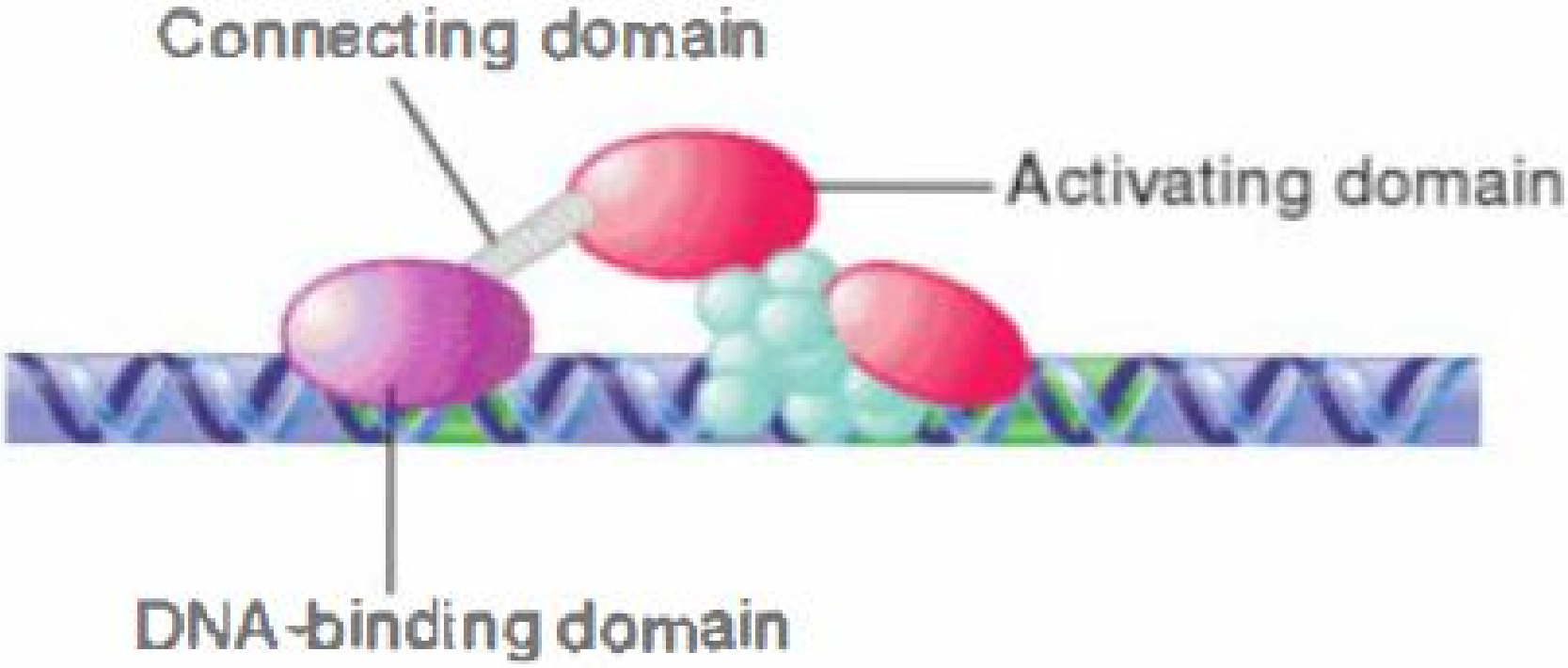


FIGURE 28.6 DNA-binding and activating functions in a transcription factor may comprise independent domains of the protein.

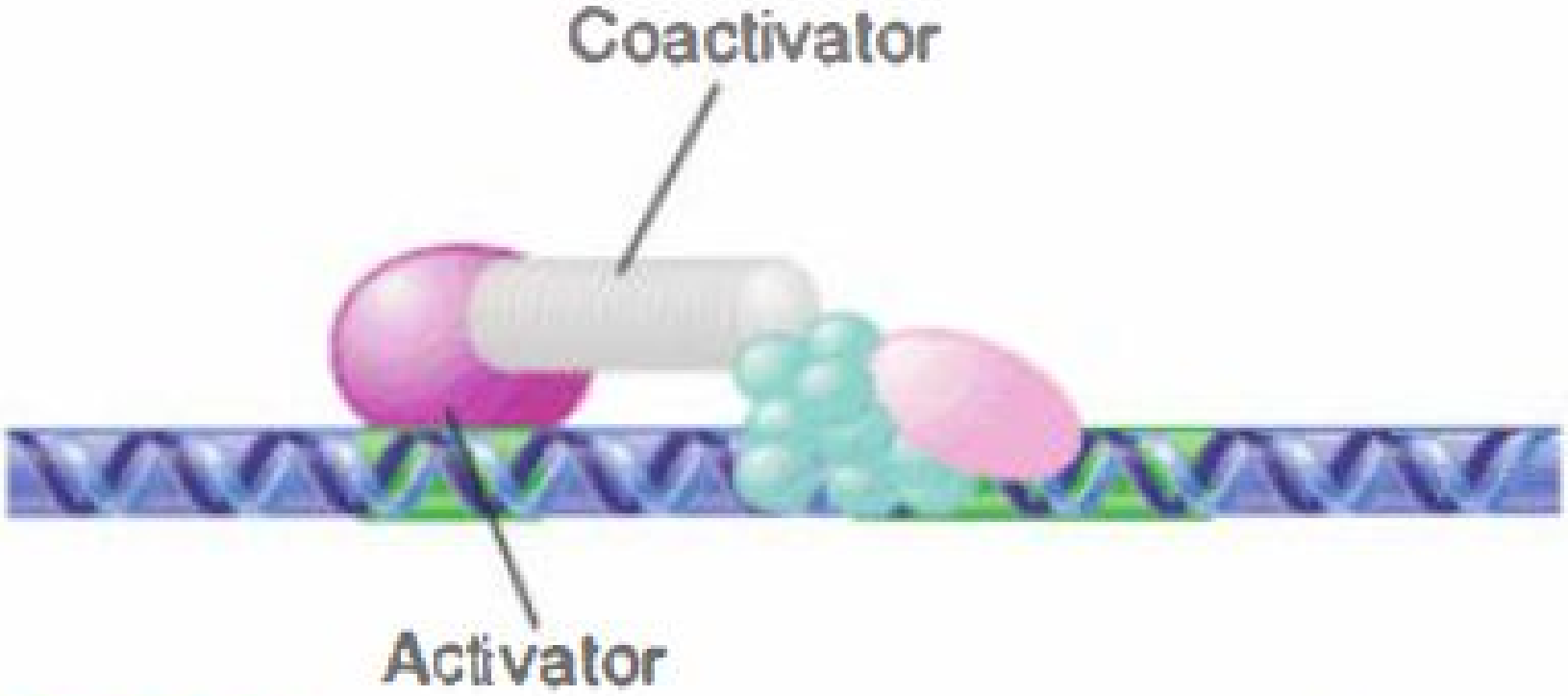


FIGURE 28.8 An activator may bind a coactivator that contacts the basal apparatus.

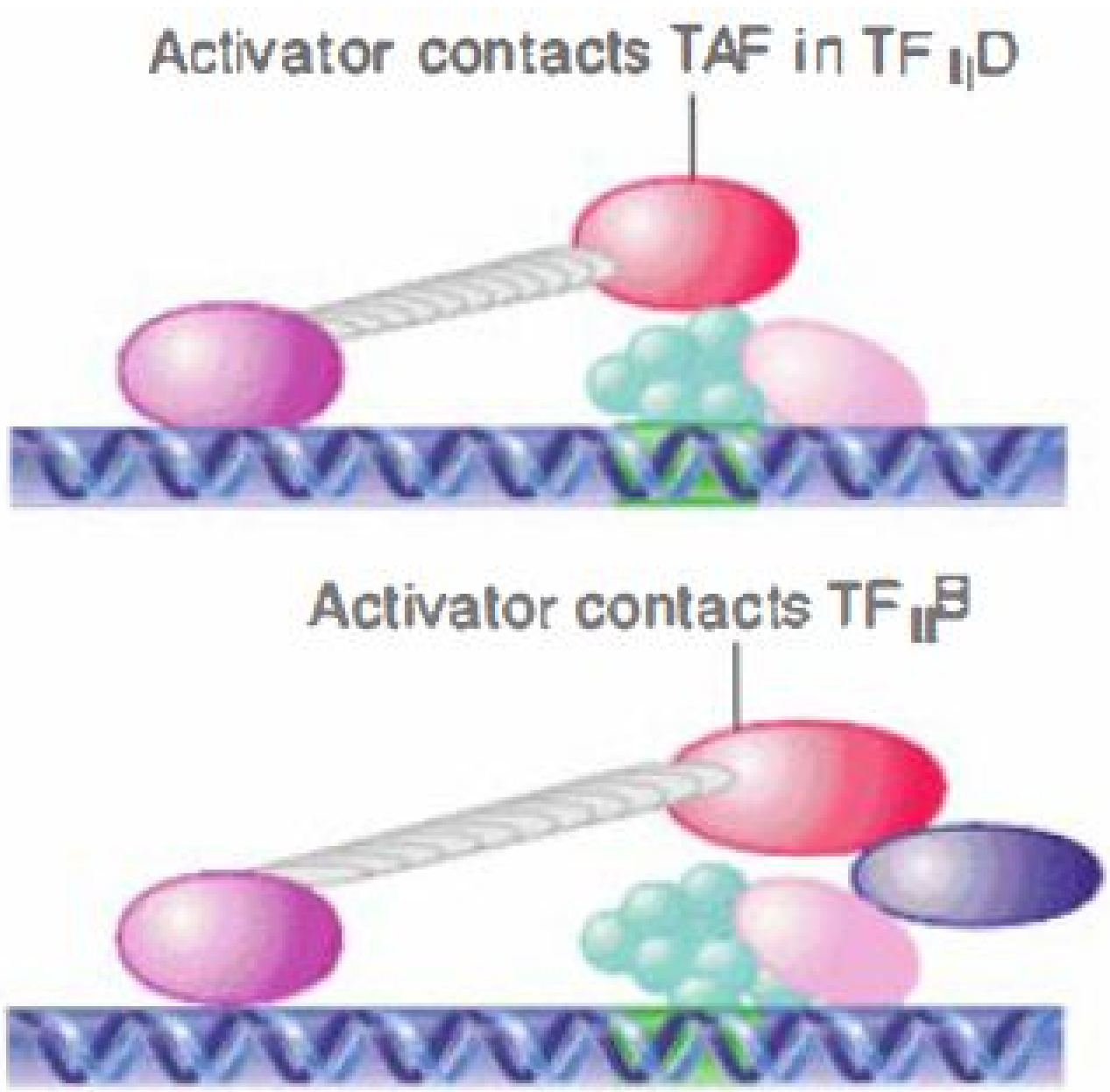


FIGURE 28.9 Activators may work at different stages of initiation bycontacting the TAFs of $TF_{II}D$ or by contacting $TF_{II}B$.

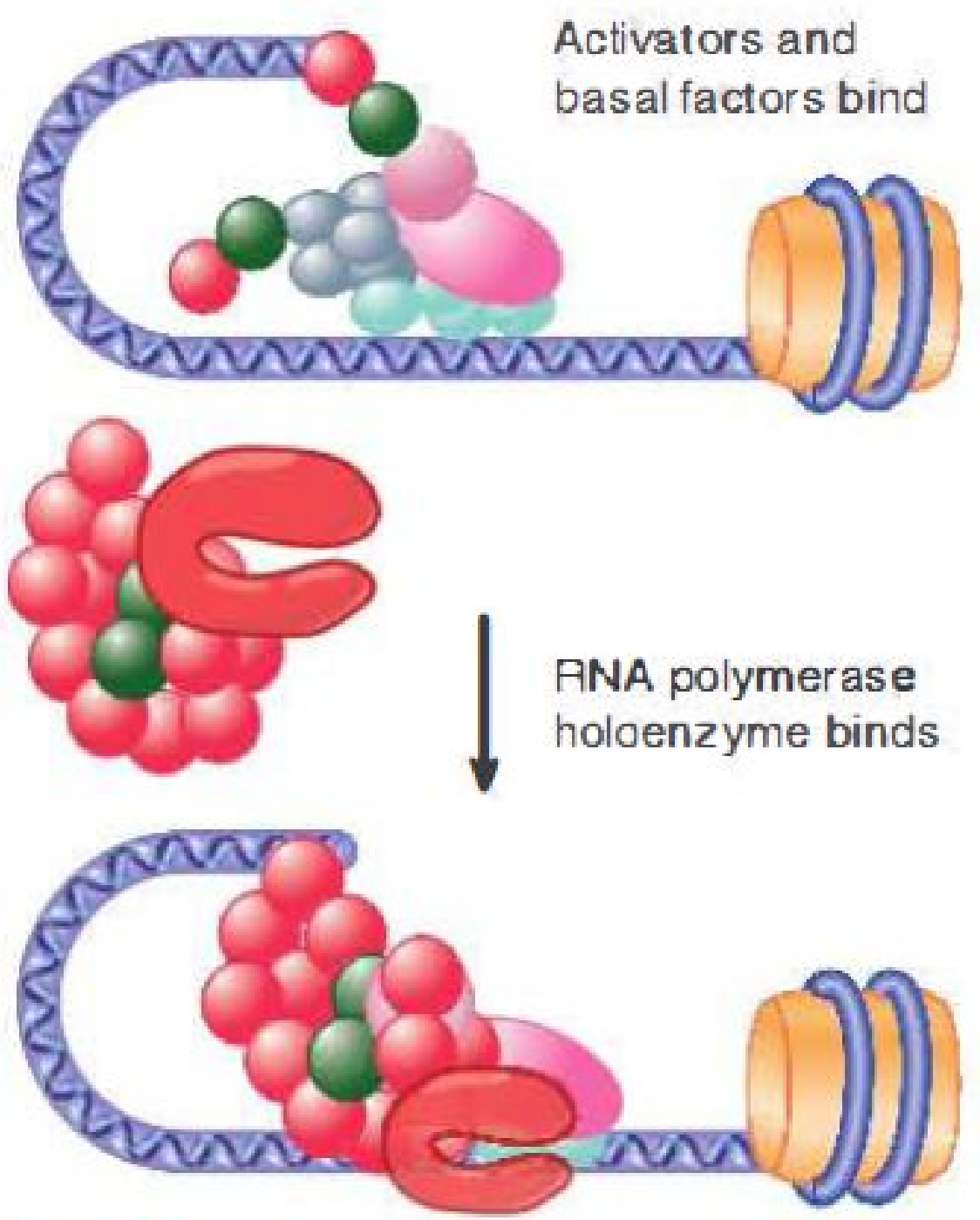
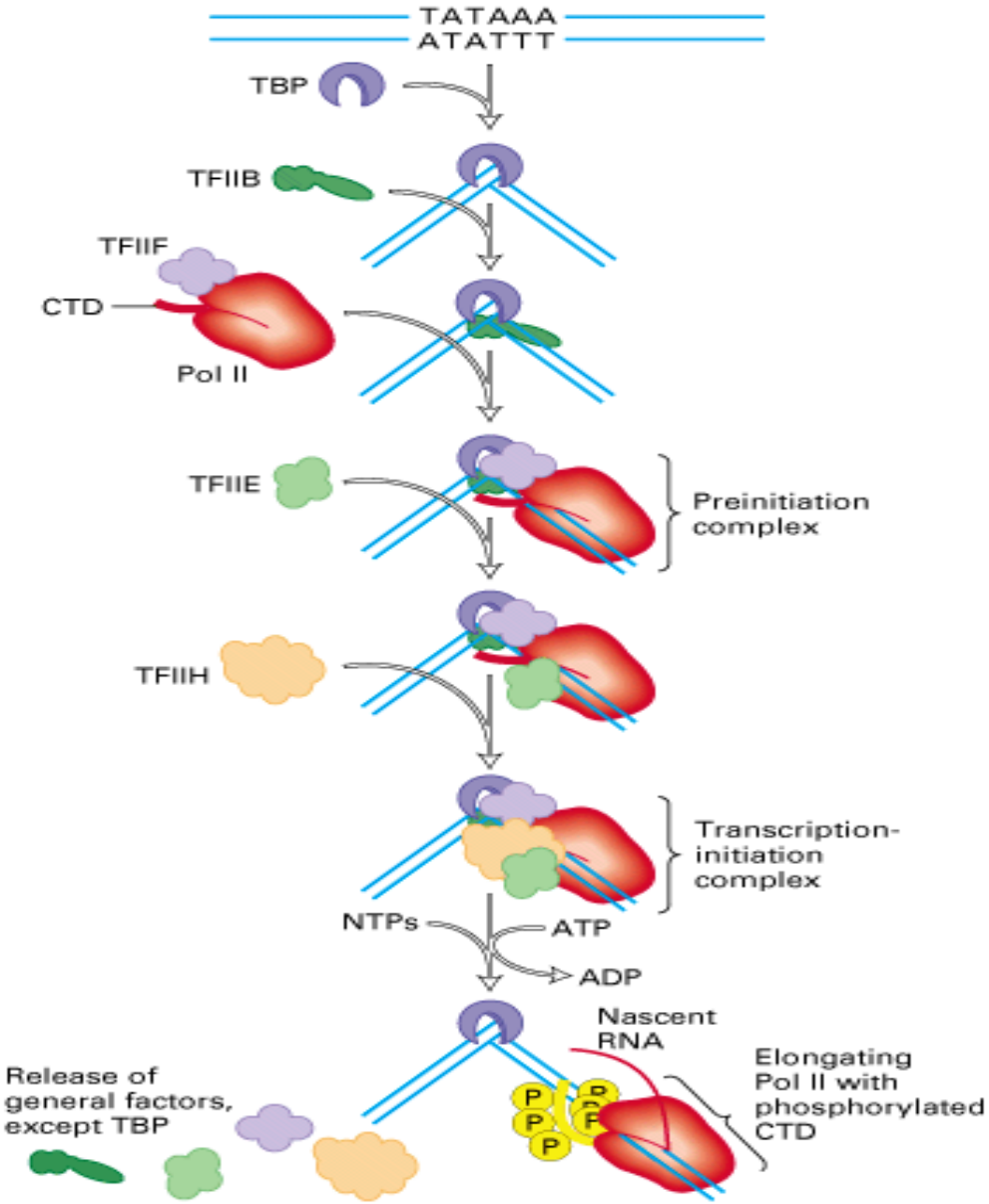
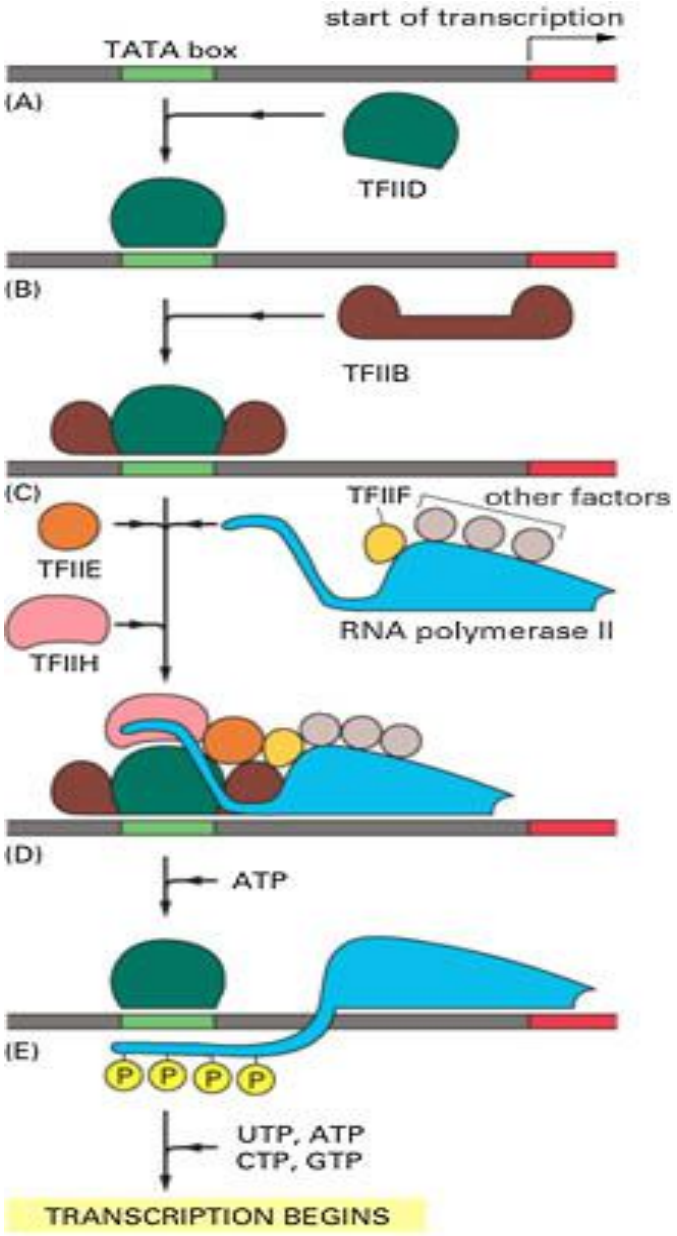


FIGURE 28.10 RNA polymerase exists as a holoenzyme containing many activators.

When the RNA polymerase starts transcribing the general factors dissociate

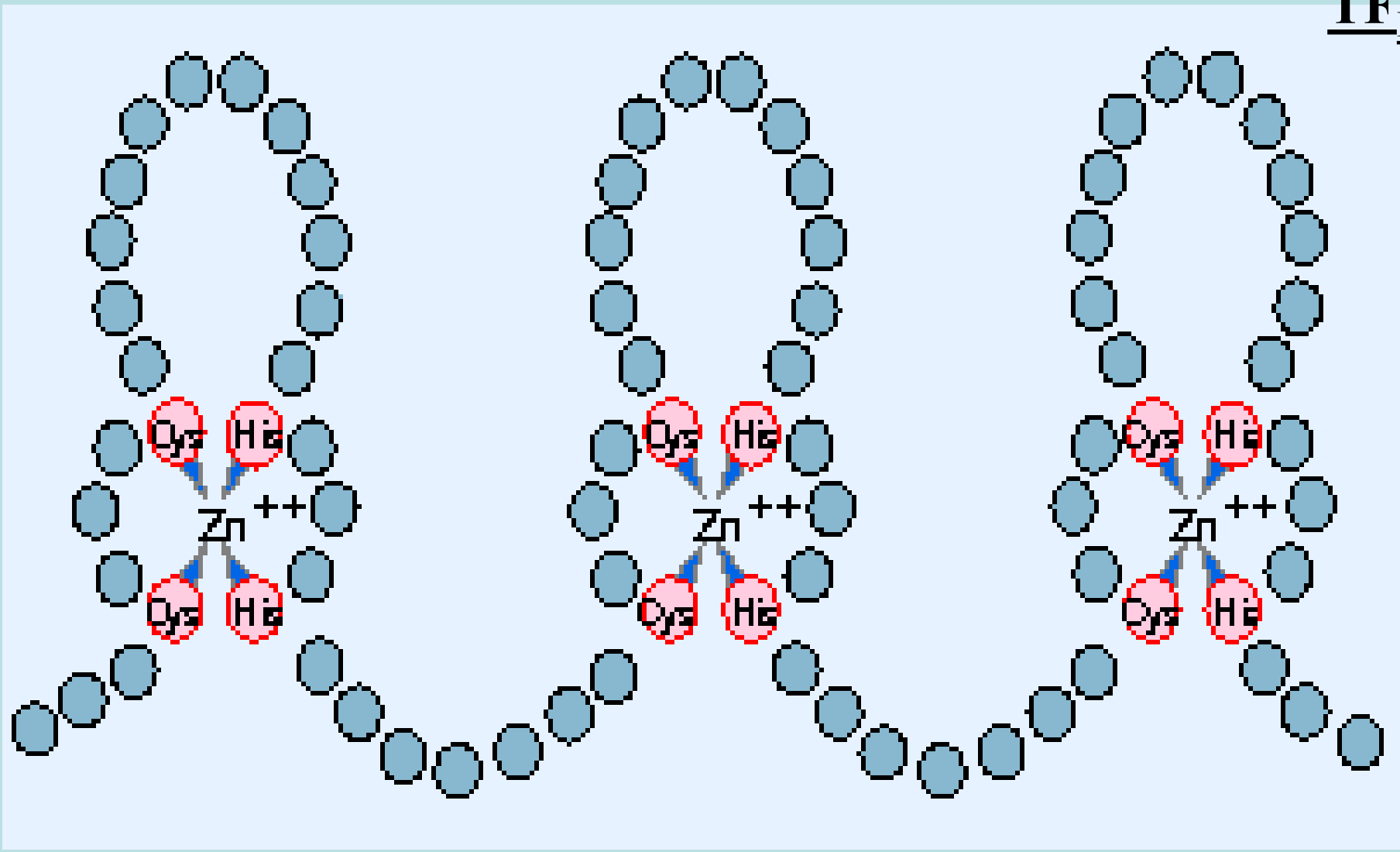




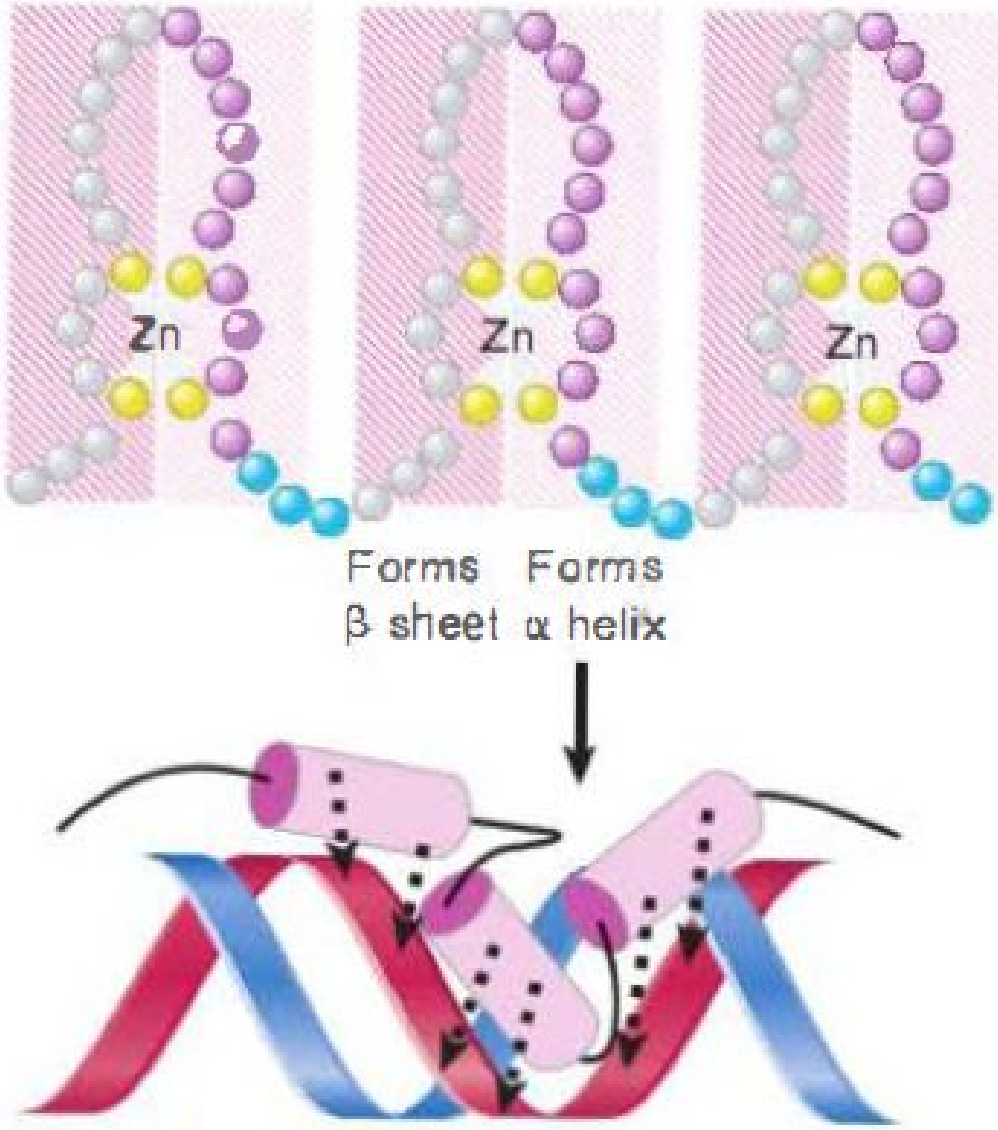
DNA binding domain structural motifs

- Zinc finger motif
- Homeodomain motif (helix-turn-helix, HTH)
- Helix-loop-helix motif (HLH)
- Leucin zipper motif

$TF_{III}A$



Transcription factor SP1 has a series of three zinc fingers, each with a characteristic pattern of cysteine and histidine residues that constitute the zinc-binding site.



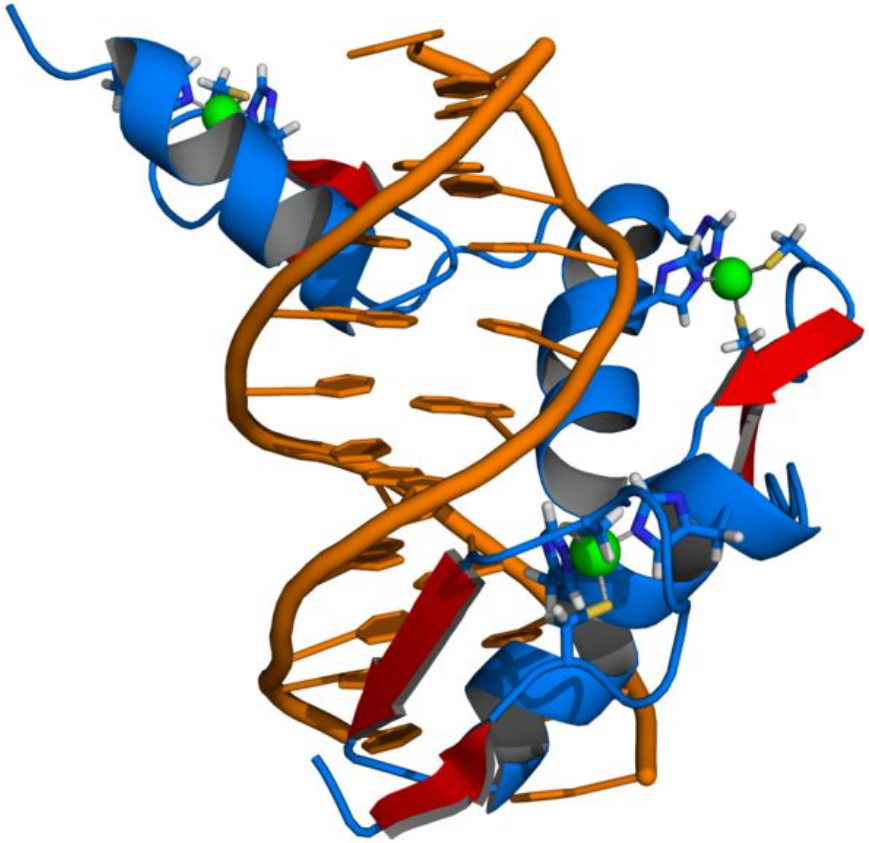


FIGURE 28.12 Zinc fingers may form α helices that insert into the major groove, which is associated with β sheets on the other side.

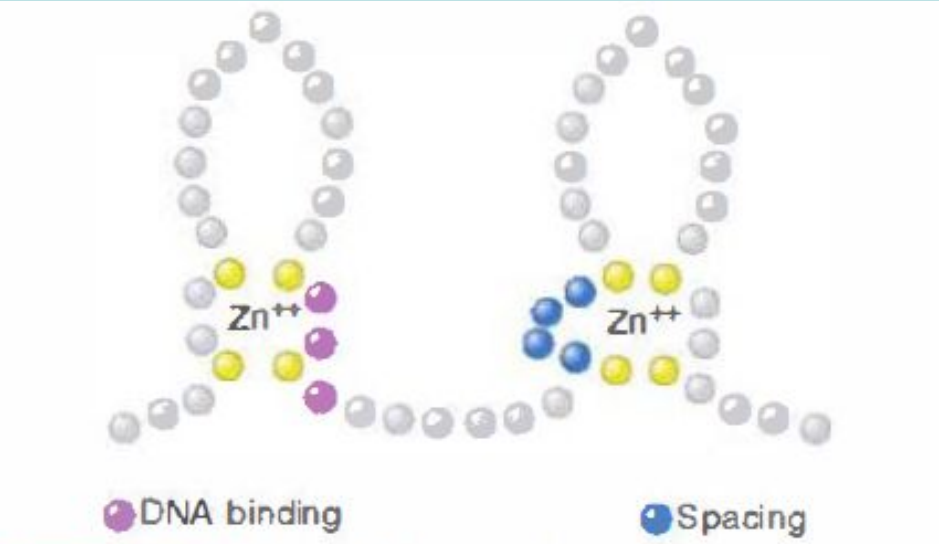
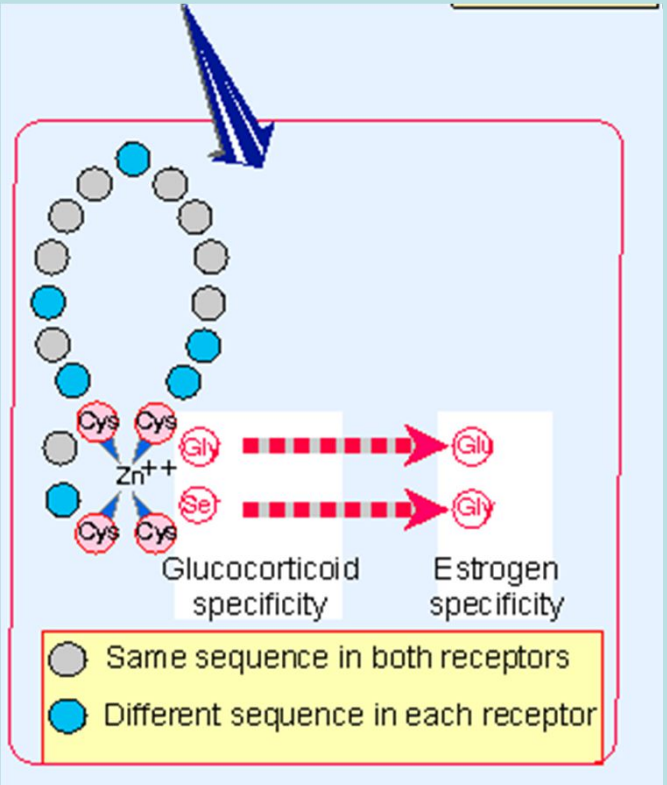


FIGURE 28.13 The first finger of a steroid receptor controls which DNA sequence is bound (positions shown in purple); the second finger controls spacing between the sequences (positions shown in blue).

Steroid receptors (Glucocorticoid and Estrogen)

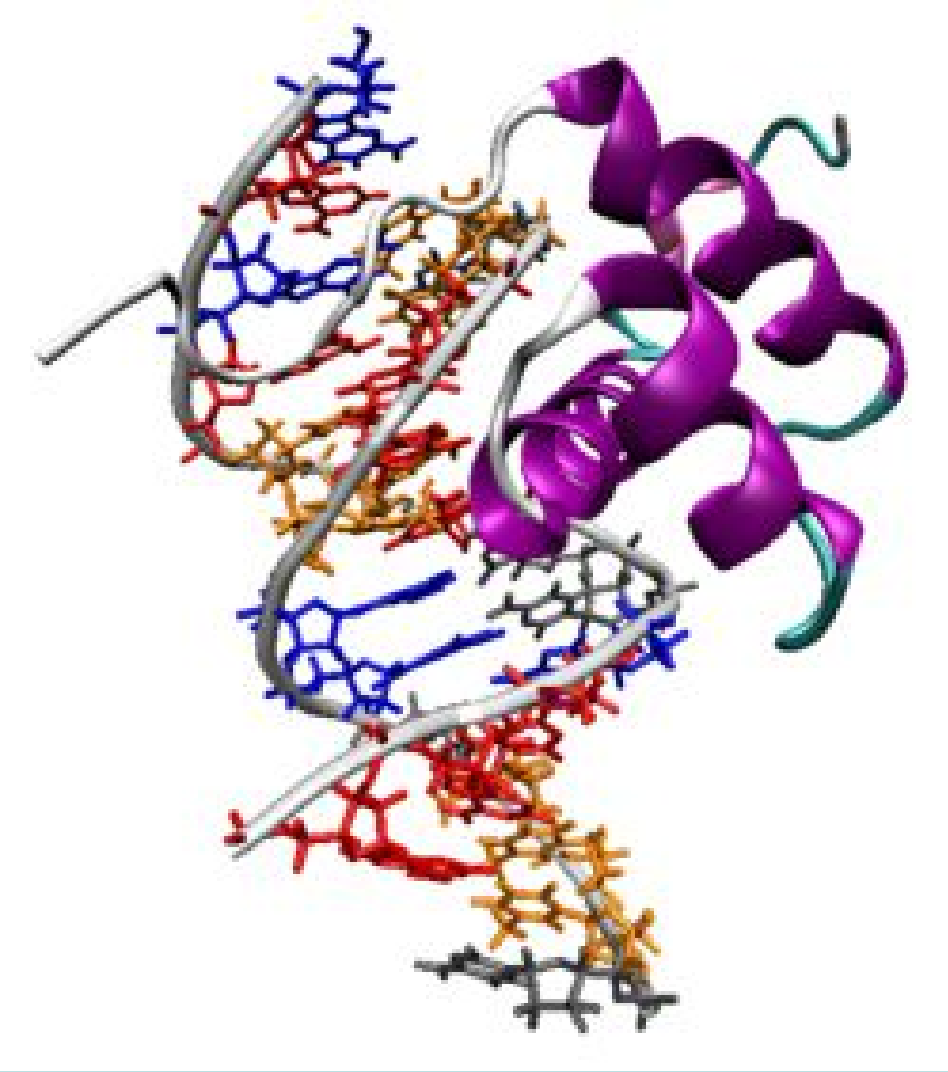


Homeodomain (Helix-Turn-Helix)

- 60 amino acids organized in 3 a helices connected by short turns.

- The third helix interacts with the major groove of the DNA target sequence.

- The N-terminal sequence interacts with the minor groove.



Helix-Loop-Helix motif (HLH)

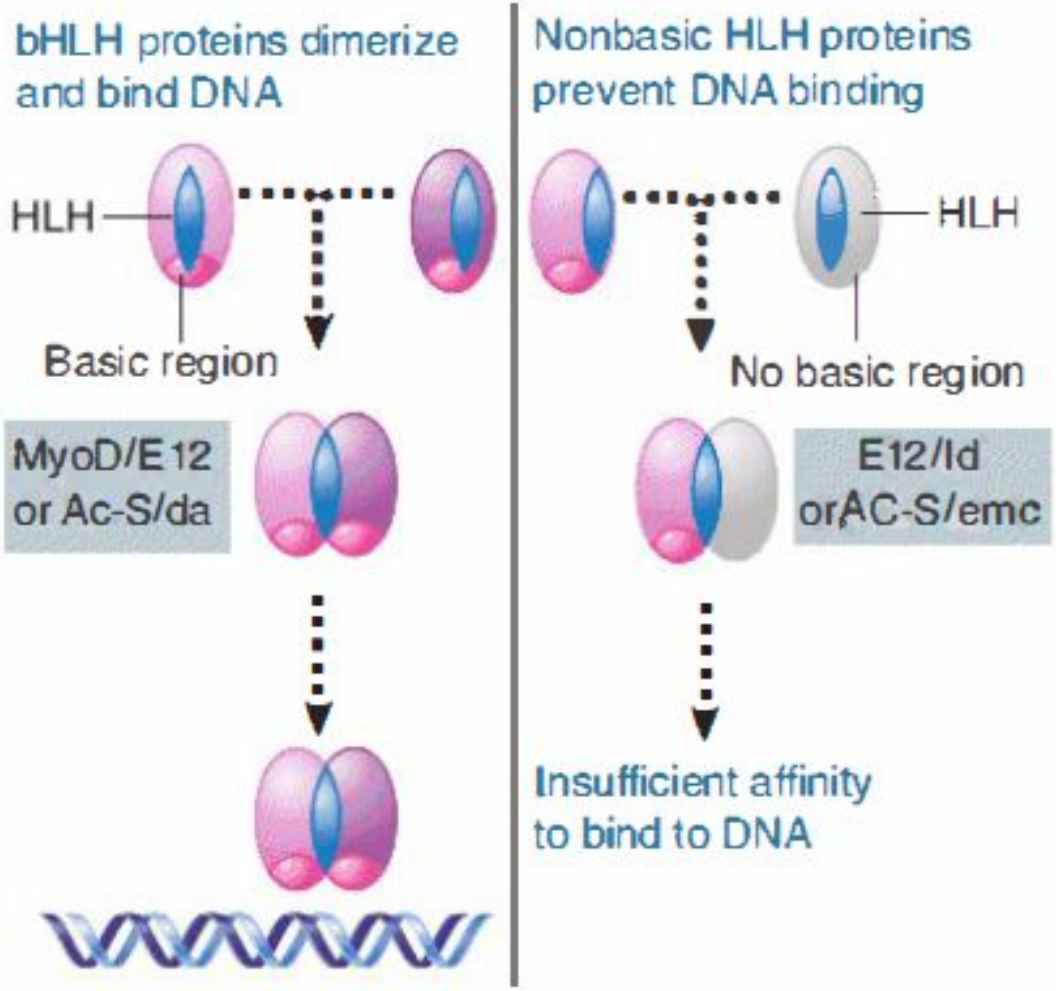
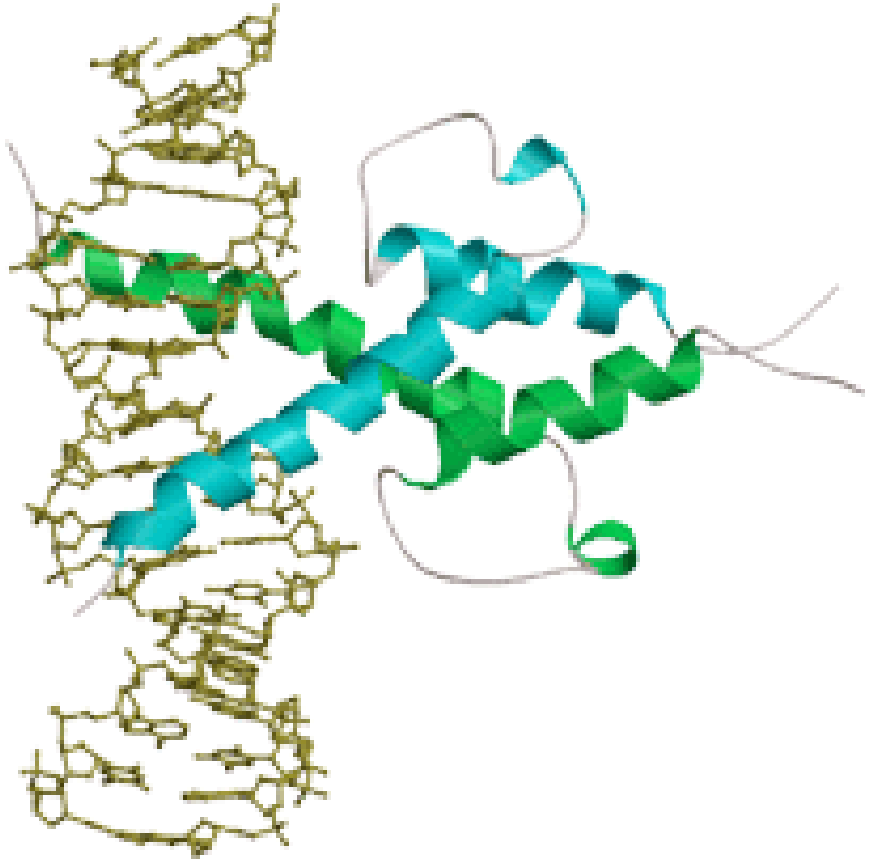


FIGURE 28.15 An HLH dimer in which both subunits are of the bHLH type can bind DNA, but a dimer in which one subunit lacks the basic region cannot bind DNA.

A Class: ubiquitary expressed
B Class : tissue specific



Leucine Zipper motif

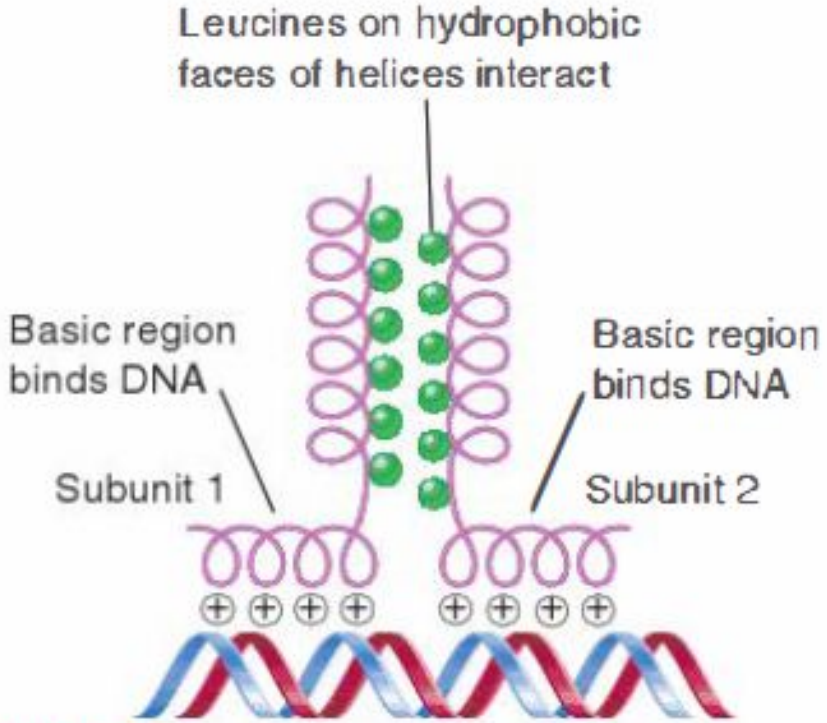
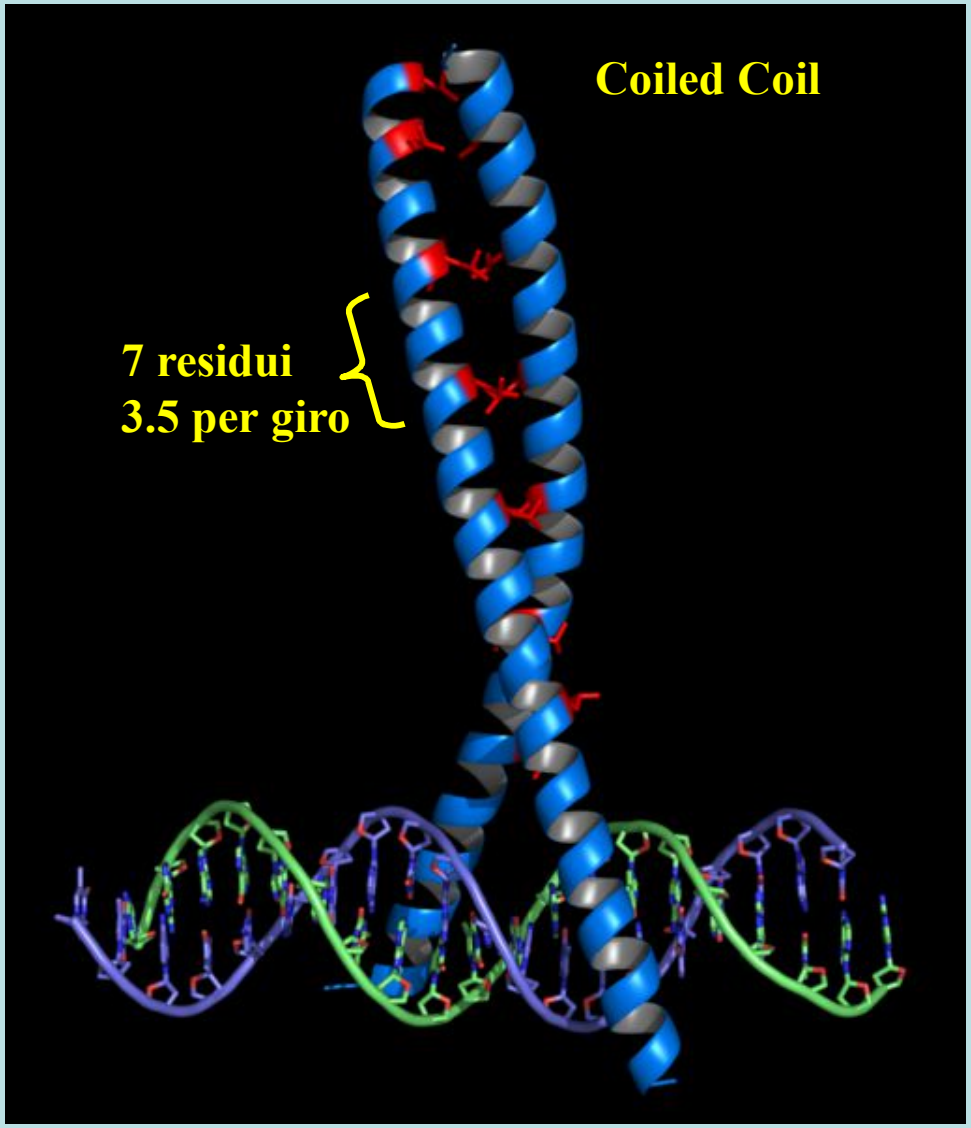
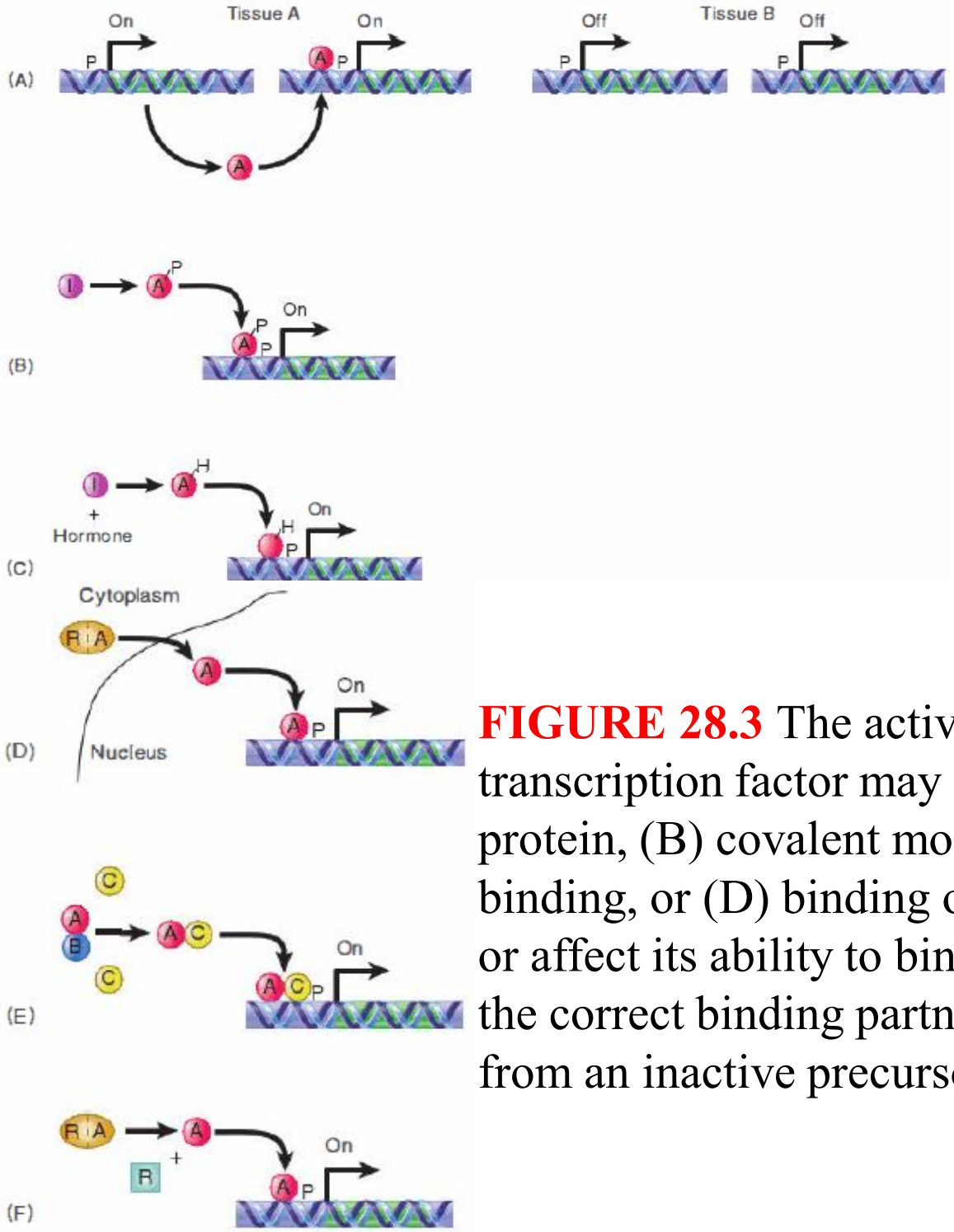


FIGURE 28.16 The basic regions of the bZIP motif are held together by the dimerization at the adjacent zipper region when the hydrophobic faces of two leucine zippers interact in parallel orientation.





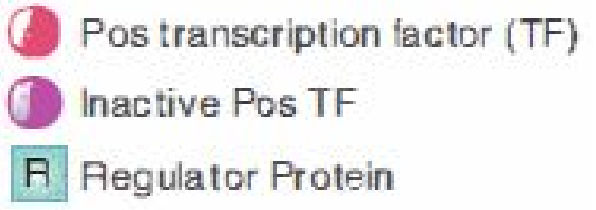
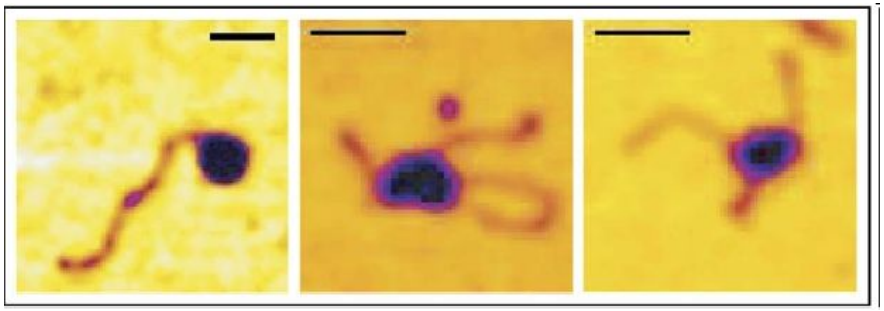


FIGURE 28.3 The activity of a positive regulatory transcription factor may be controlled by (A) synthesis of protein, (B) covalent modification of protein, (C) ligand binding, or (D) binding of inhibitors that sequester the protein or affect its ability to bind to DNA {E) by the ability to select the correct binding partner for activation, and (F) by cleavage from an inactive precursor.



Type of Complex	SWI/SNF	ISWI	CHD	INO80/SWRI
Yeast	SWI/SNF RSC	ISW1a, ISWb ISW2	CHDI	INO80 SWRI
Fly	dSWI/SNF (brahma)	NURF CHRAC ACF	JMIZ	Tip60
Human	hSWI/SNF	RSF hACF/WCFR hCHRAC WICH	NuRD	INO80 SRCAP
Frog		WICH CHRAC ACF	Mi-2	

FIGURE 28.20 Remodeling complexes can be classified by their ATPase subunits.

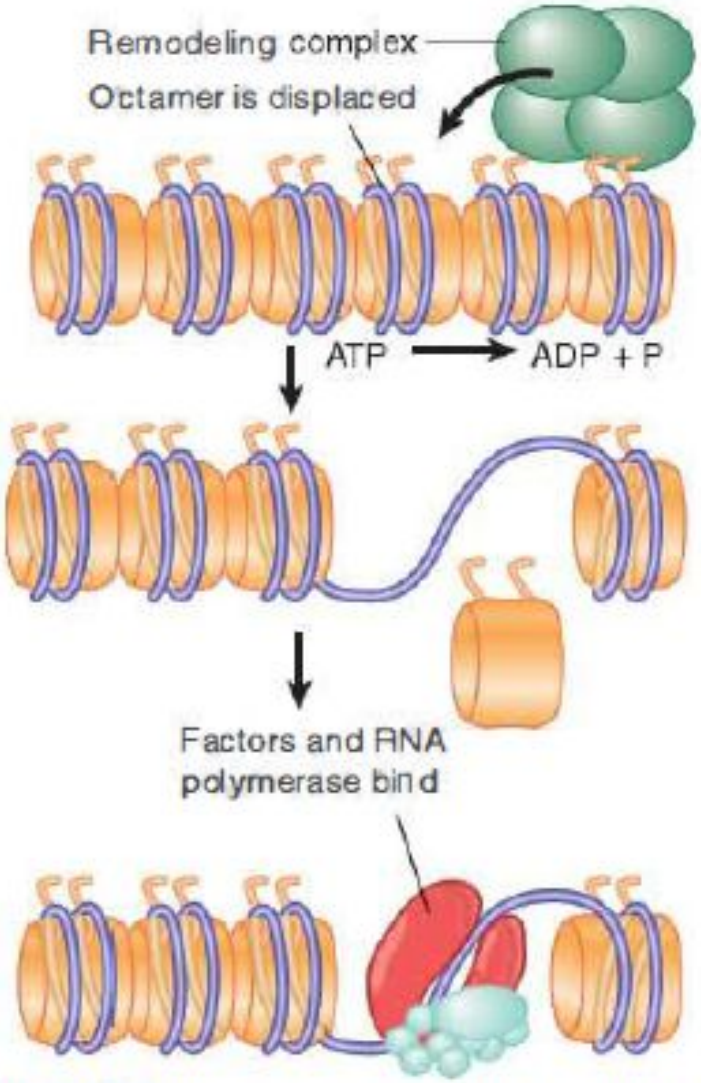


FIGURE 28.18 The dynamic model for transcription of chromatin relies upon factors that can use energy provided by hydrolysis of ATP to displace nucleosomes from specific DNA sequences.

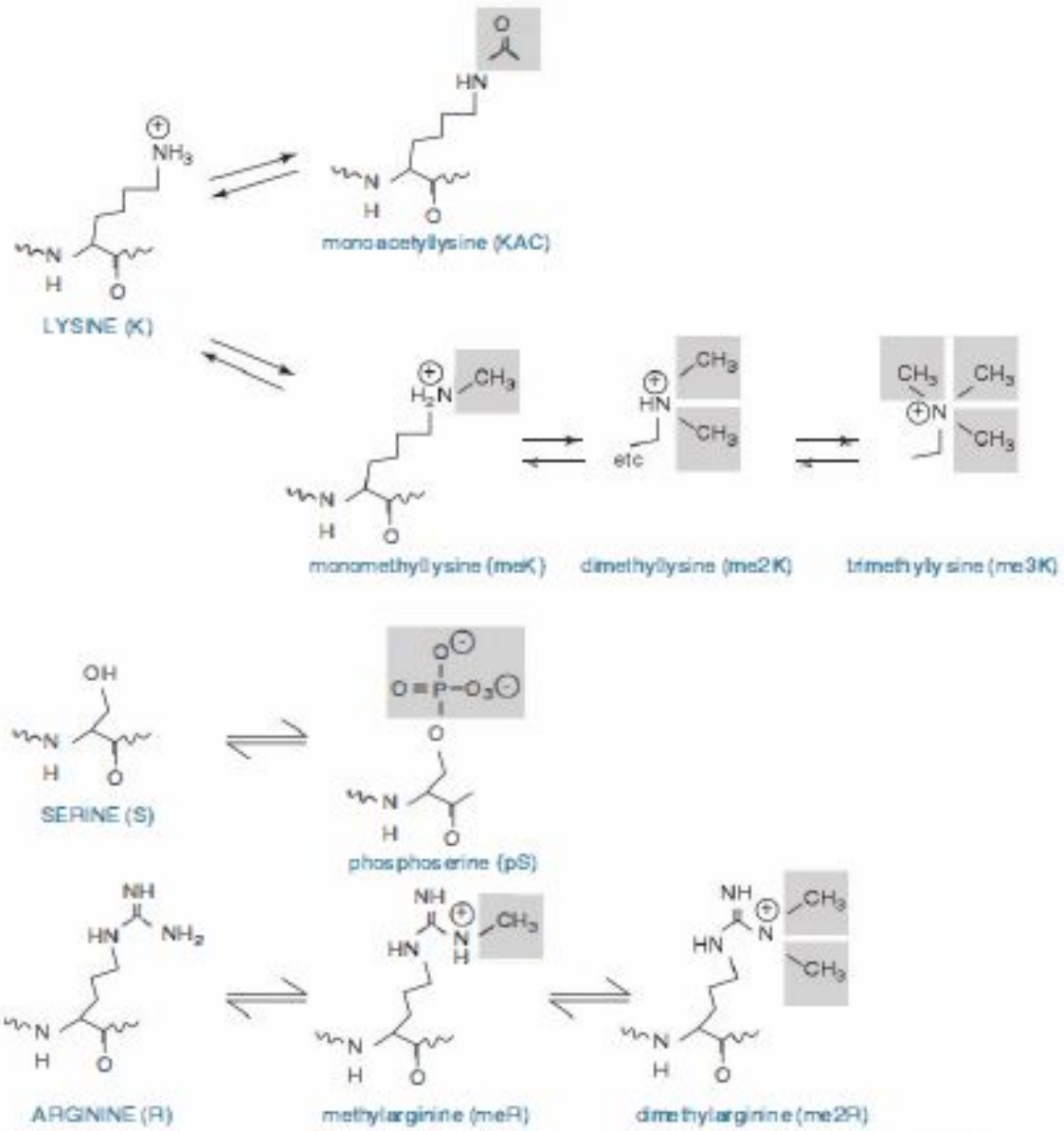


FIGURE 10.17 The positive charge on lysine is neutralized upon acetylation, while methylated lysine and arginine retain their positive charges. Lysine can be mono-, di-, or triacetylated, while arginine can be mono- or diacetylated. Serine or threonine phosphorylation results in a negative charge.

Histone	Site	Modification	Function
H3	K-4	Methylation	Transcription activation
H3	K-9	Methylation	Chromatin condensation
	K-9	Methylation	Required for DNA methylation
	K-9	Acetylation	Transcription activation
H3	S-10	Phosphorylation	Transcription activation
H3	K-14	Acetylation	Prevents methylation at Lys-9
H3	K-79	Methylation	Telomeric silencing
H4	R-3	Methylation	Transcription regulation
H4	K-5	Acetylation	Nucleosome assembly
H4	K-12	Acetylation	Nucleosome assembly
H4	K-16	Acetylation	Nucleosome assembly, fly X activation

FIGURE 10.20 Most modified sites in histones have a single, specific type of modification, but some sites can have more than one type of modification. Individual functions can be associated with some of the modifications.

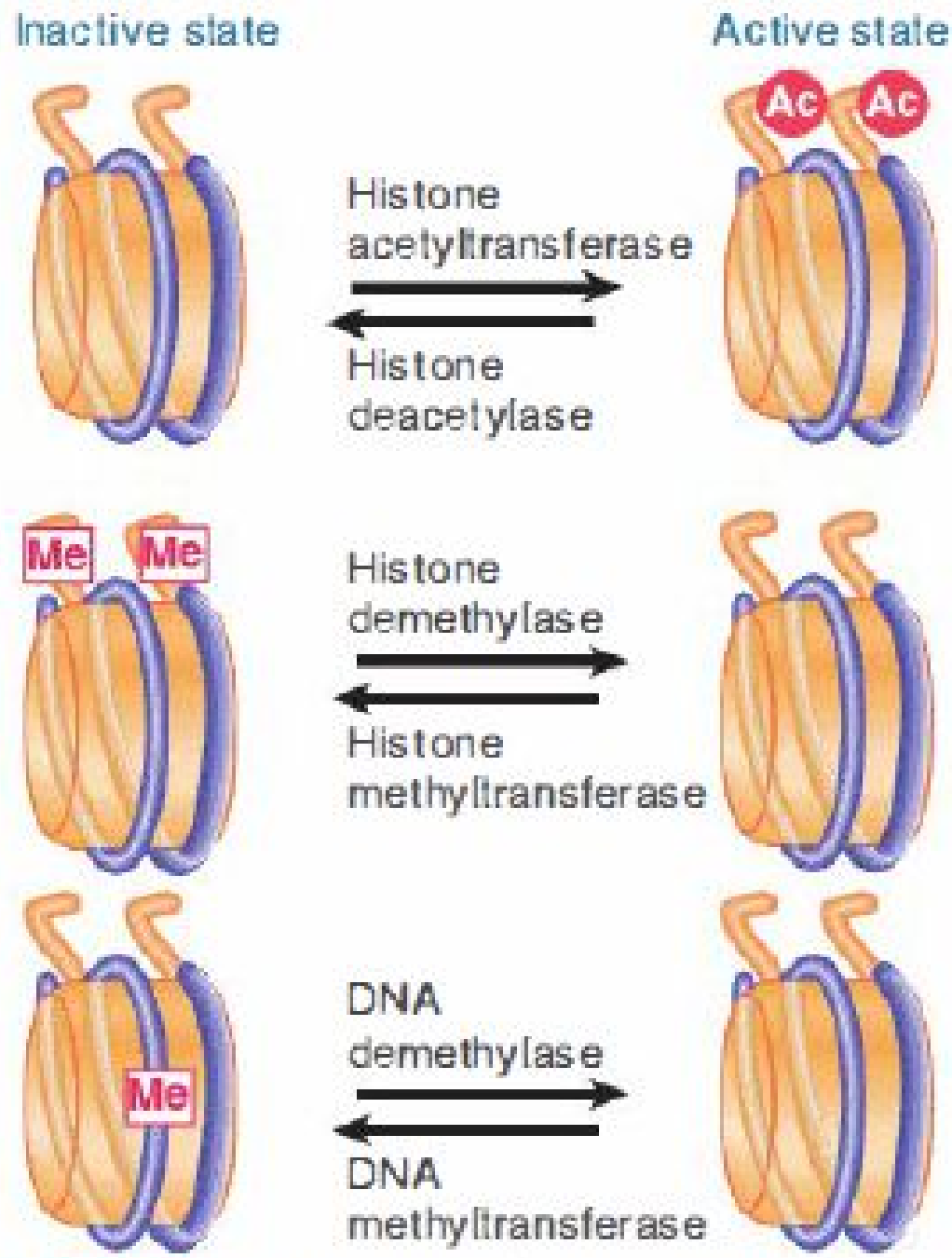


FIGURE 28.28 Acetylation of histones activates chromatin, and methylation of DNA and specific sites on histones inactivates chromatin.

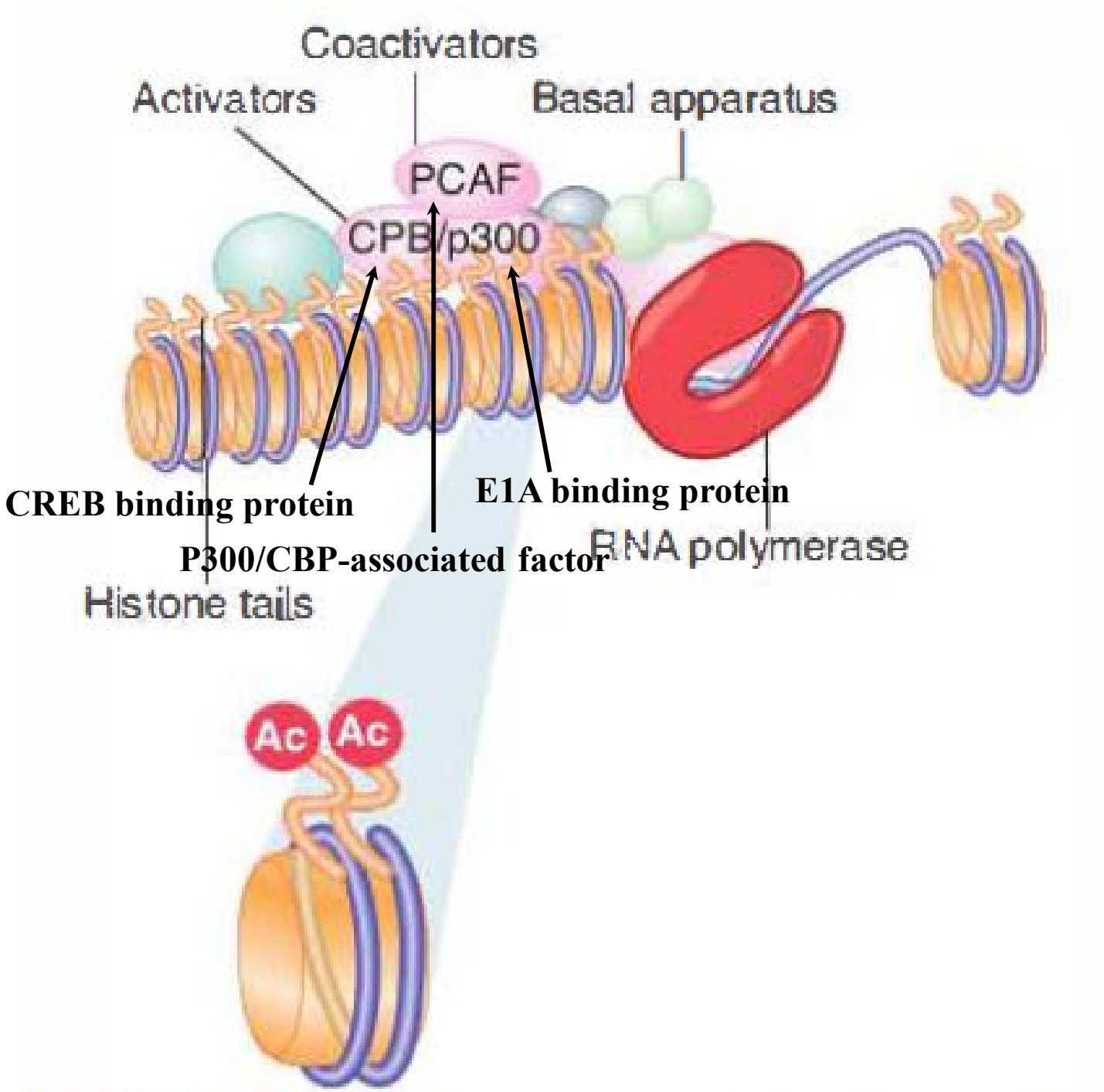


FIGURE 28.24 Coactivators may have HAT activities that acetylate the tails of nucleosomal histones.

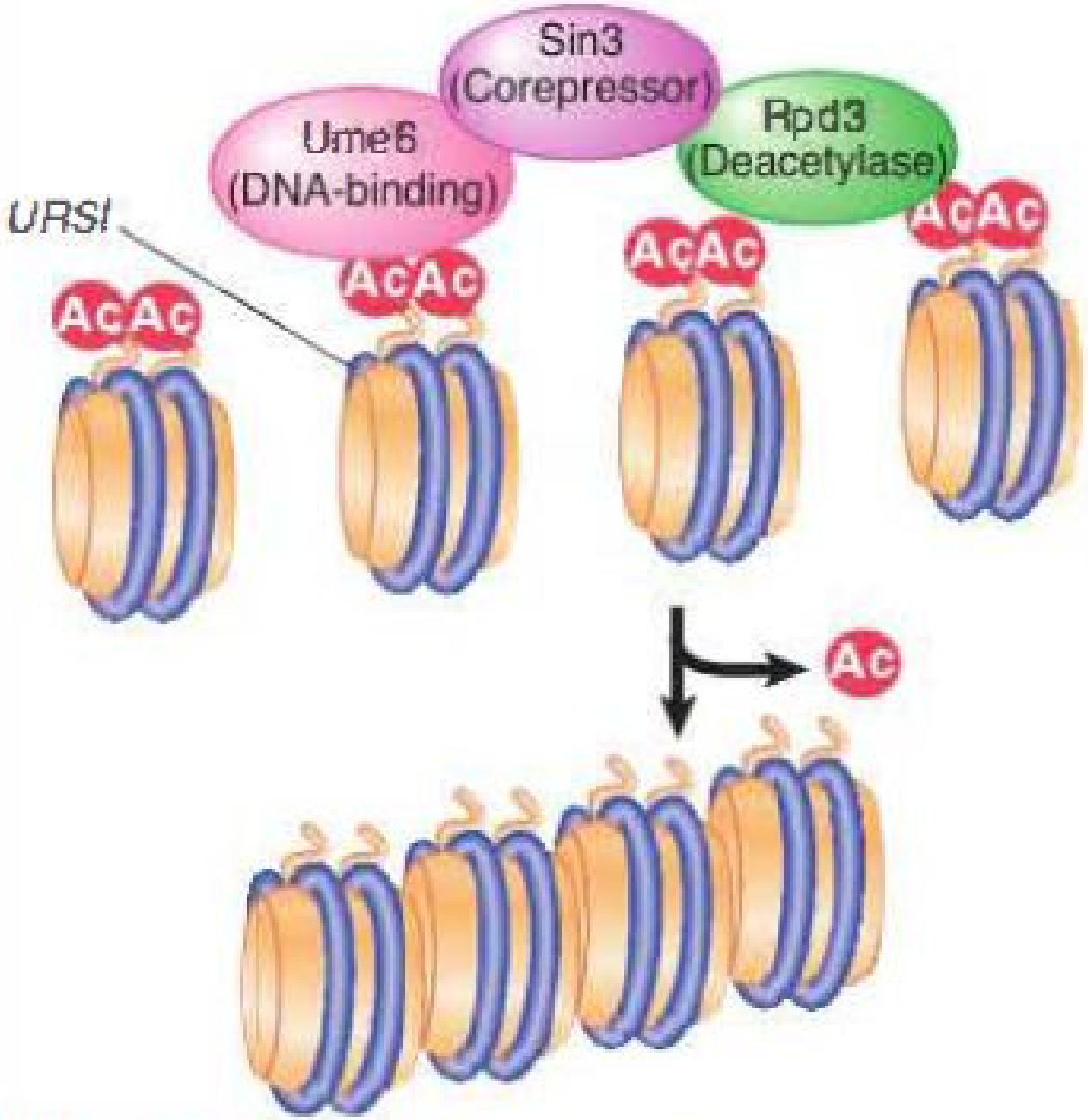
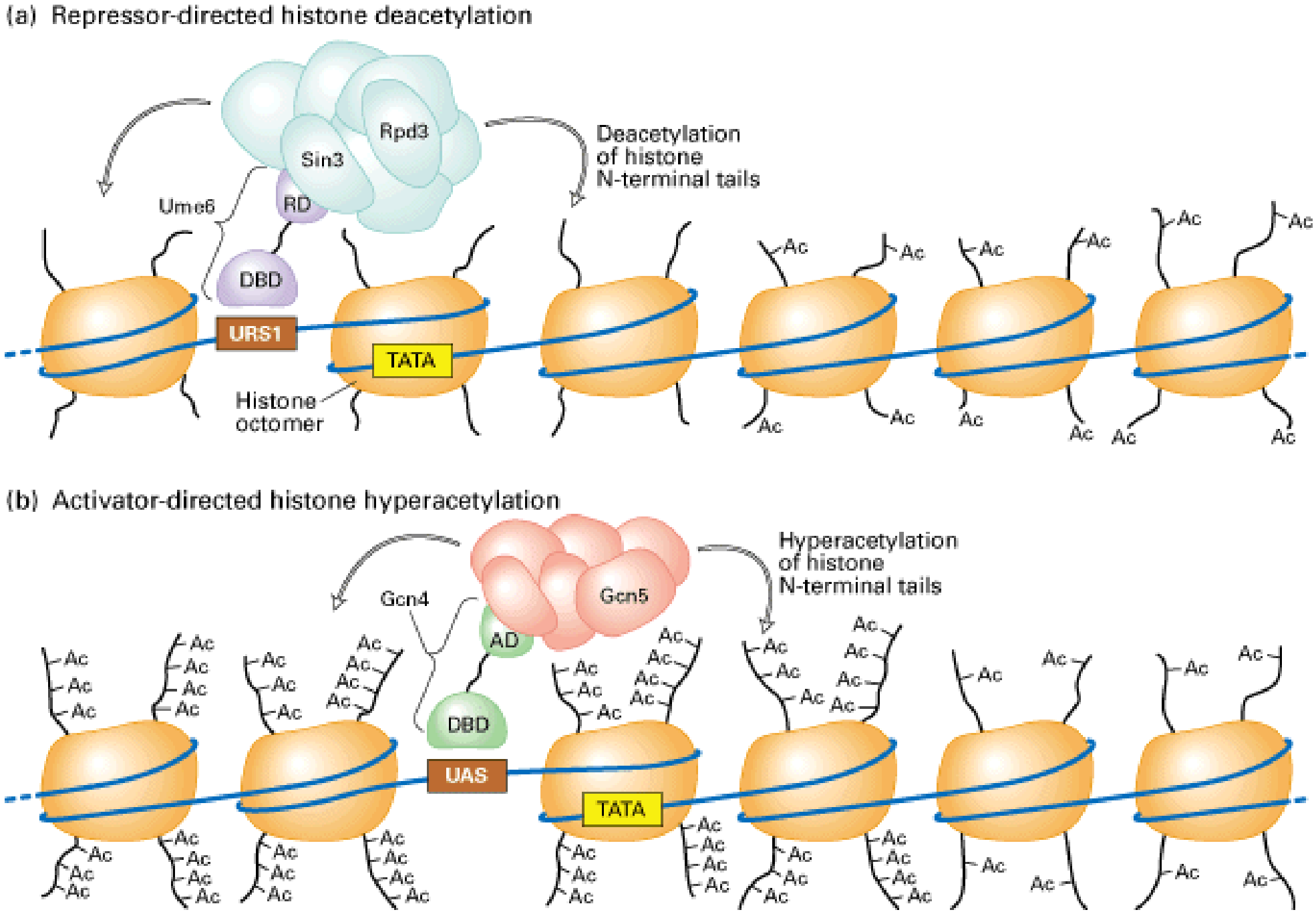


FIGURE 28.26 A repressor complex contains three components: a DNA-binding subunit, a corepressor, and a histone deacetylase.


(a) Repressor-directed histone deacetylation
Rpd3
Sin3
RD
Ume6
DBD
URS1
TATA
Histone
octomer
Deacetylation
of histone
N-terminal tails
Ac
(b) Activator-directed histone hyperacetylation
Gcn5
Gcn4
AD
DBD
UAS
TATA
Hyperacetylation
of histone
N-terminal tails
Ac

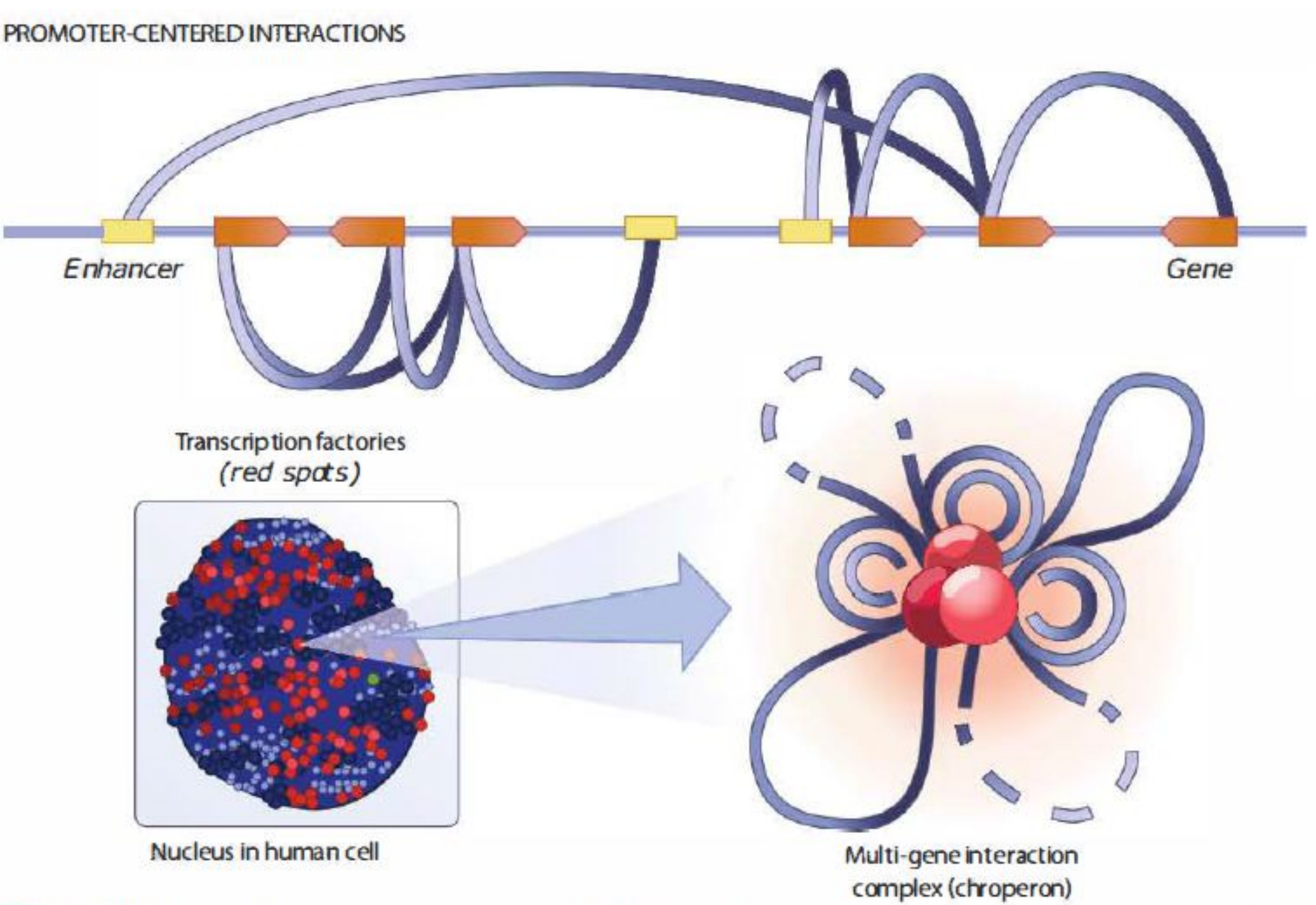
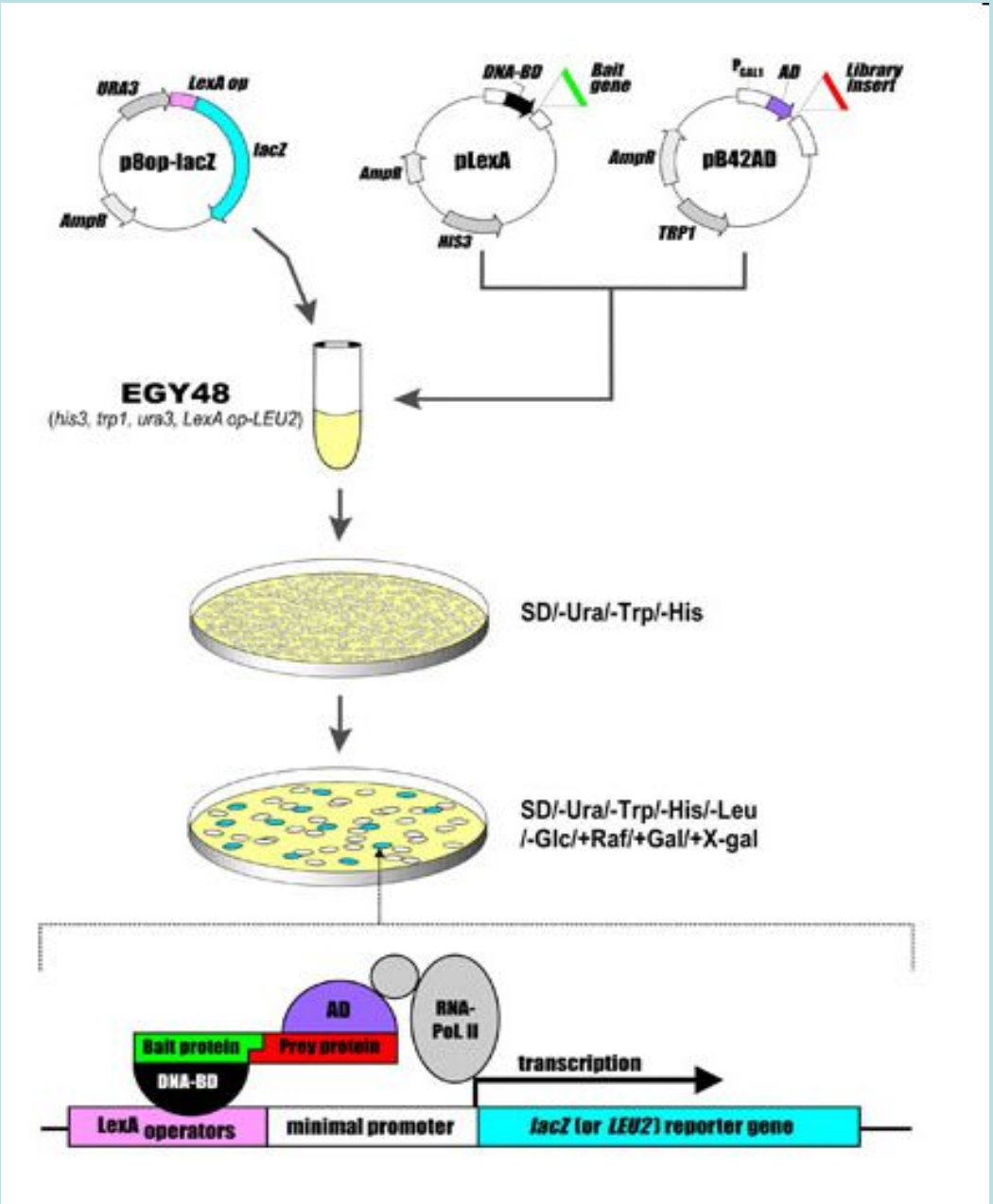
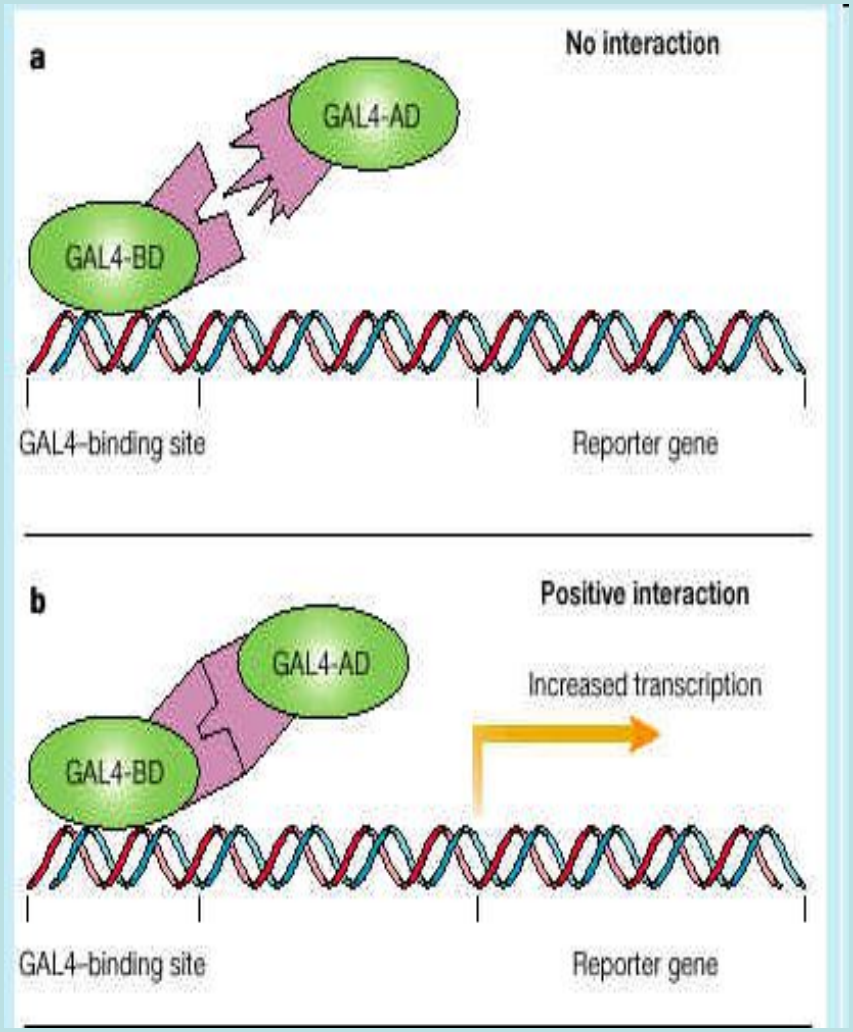


FIGURE 28.11 Higher order chromatin interactions synergistically promote transcription of clustered genes. These interactions indicate a topological, combinatorial mechanism of transcription regulation. Modified from *Cell* 148 (2012): 1-7.

(Yeast) Two Hybrid System





(Yeast) Two Hybrid System

- ***In vivo*** **study of the interaction between two proteins (or other macromolecules).**

- **Screening of different interactors (for example a gene library) to find possible partners of interaction for a specific protein.**