Regulation of gene expression

(Lehninger pg. 1072 - 1085)

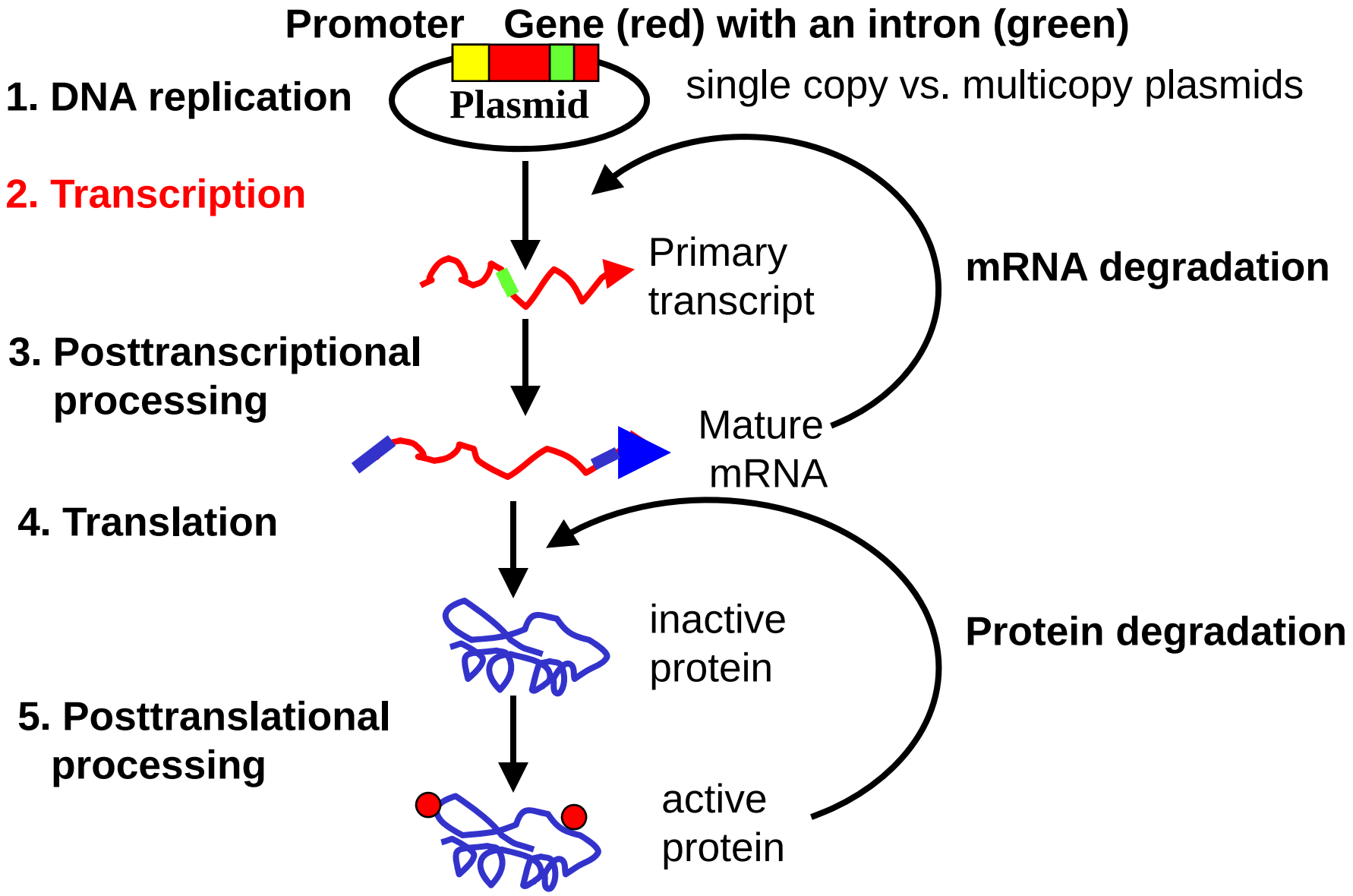
Today’s lecture

- Gene expression
- Constitutive, inducible, repressible genes
- Specificity factors, activators, repressors
- Negative and positive gene regulation
- Lac operon
- Helix-turn-helix motifs
- Zinc-fingers
- Leucine zippers

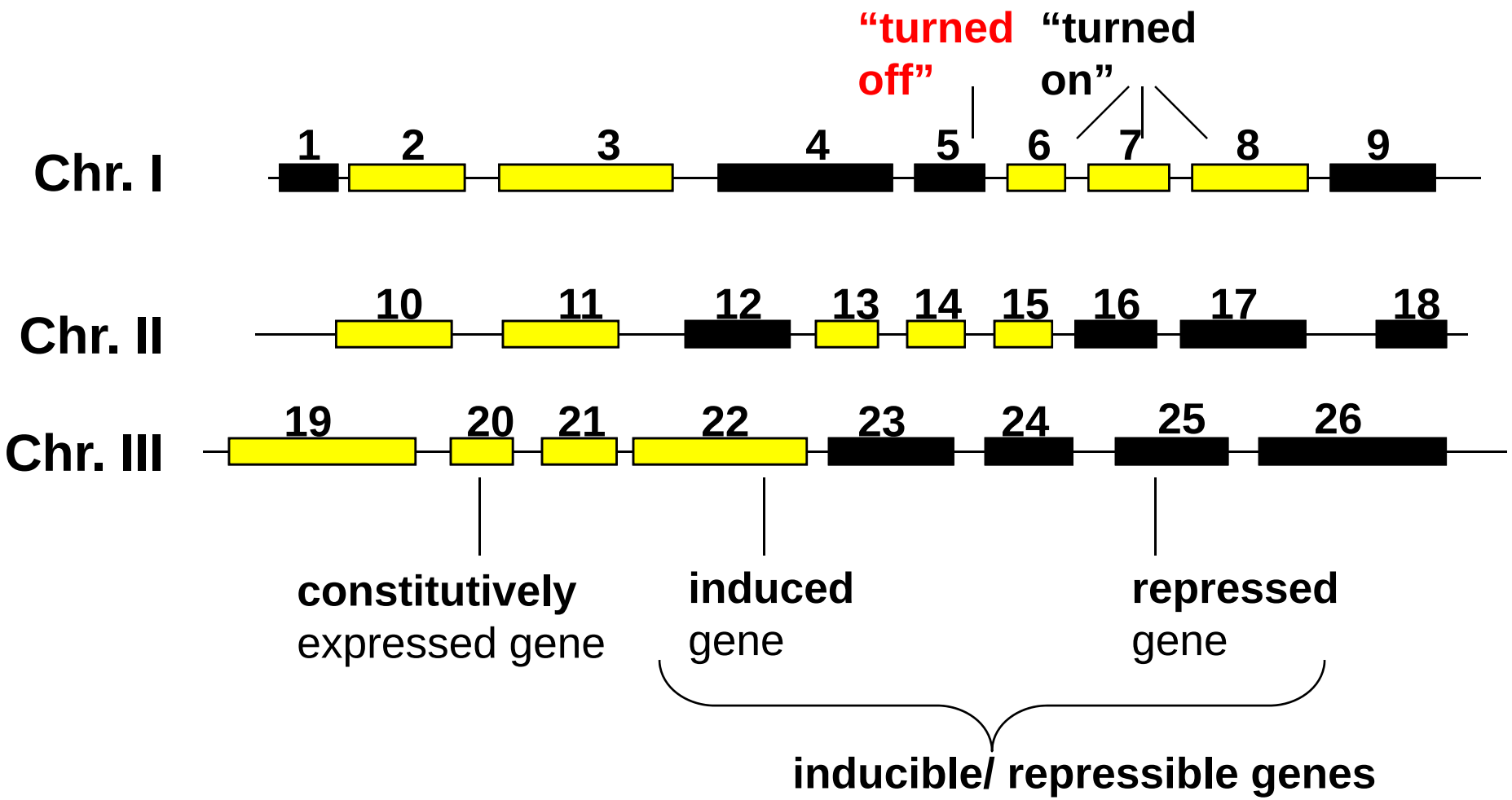
What is gene expression?

- Biological processes, such as transcription, and in case of proteins, also translation, that yield a gene product.
- A gene is expressed when its biological product is present and active.
- Gene expression is regulated at multiple levels.

Regulation of gene expression

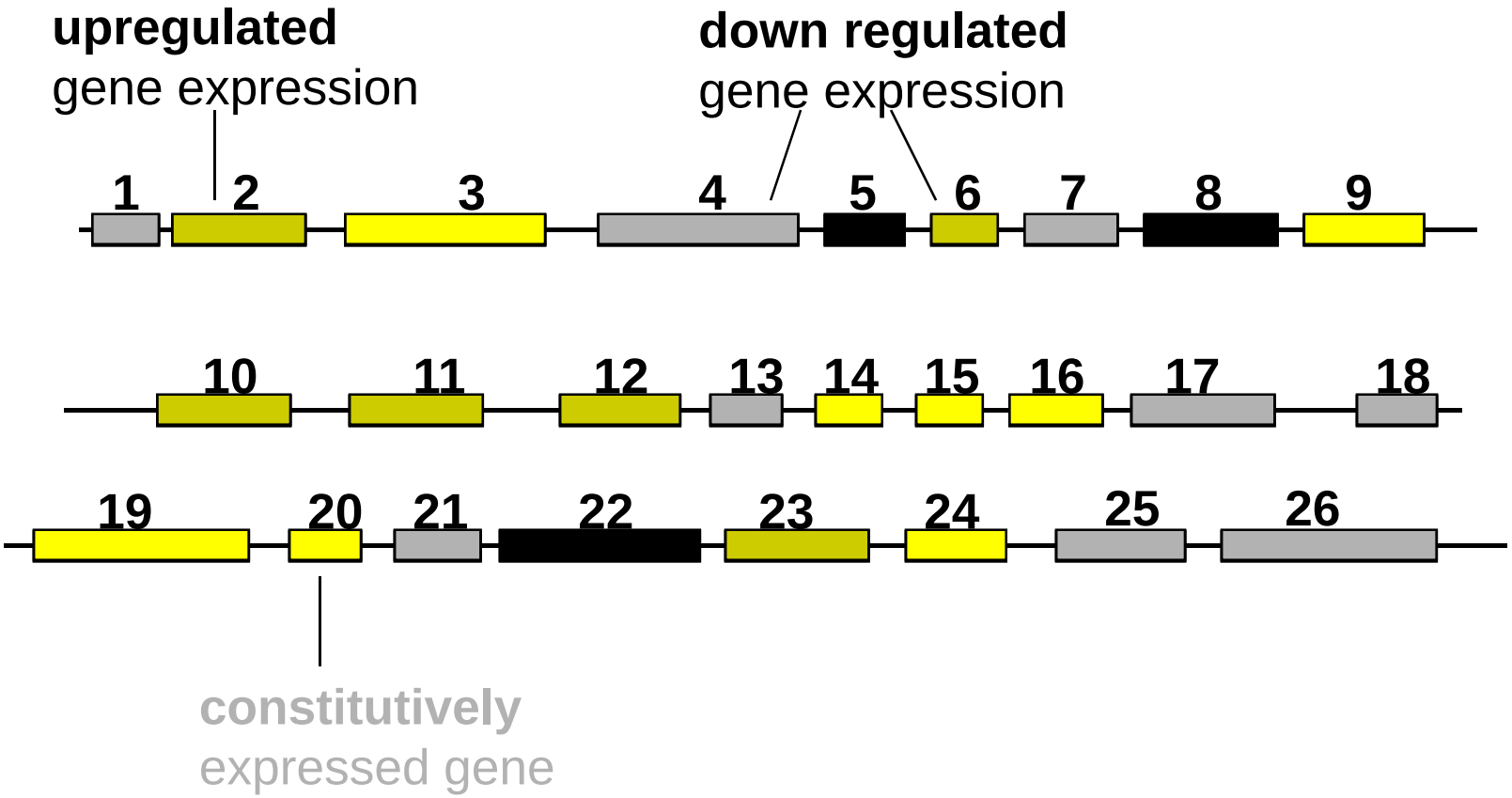


Gene regulation (1)



Gene regulation (2)

Condition 4



Definitions

- Constitutively expressed genes:
  - Genes that are actively transcribed (and translated) under all experimental conditions, at essentially all developmental stages, or in virtually all cells.
- Inducible genes:
  - Genes that are transcribed and translated at higher levels in response to an inducing factor
- Repressible genes:
  - Genes whose transcription and translation decreases in response to a repressing signal

Definitions

- Housekeeping genes:
  - genes for enzymes of central metabolic pathways (e.g. TCA cycle)
  - these genes are constitutively expressed
  - the level of gene expression may vary

Modulators of transcription

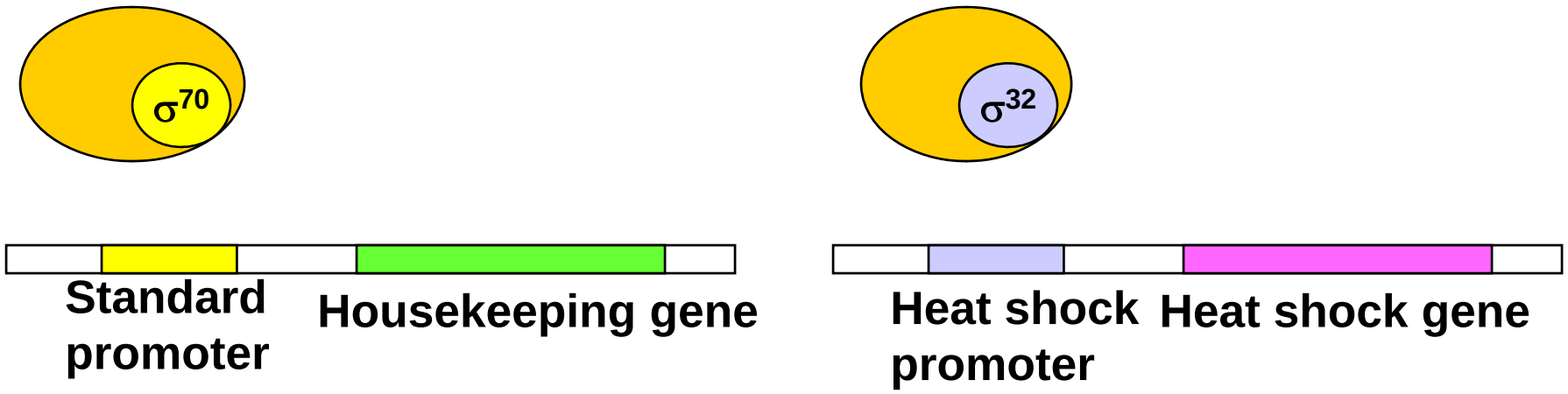
- Modulators:

  (1) specificity factors, (2) repressors, (3) activators

1. Specificity factors:

   Alter the specificity of RNA polymerase

   Examples: σ-factors (σ^{70}, σ^{32}), TBPs



Modulators of transcription

2. Repressors:

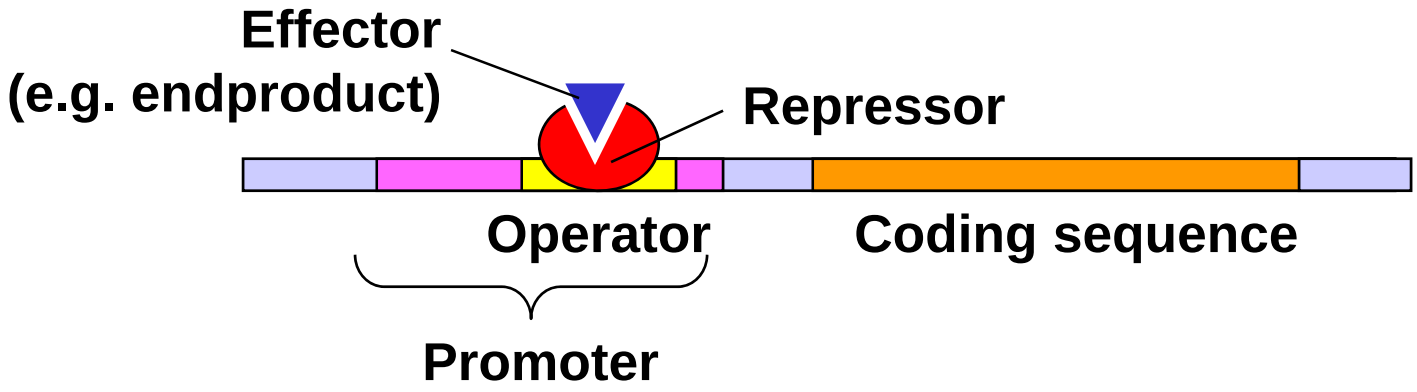
   mediate **negative gene regulation**

   may impede access of RNA polymerase to the promoter

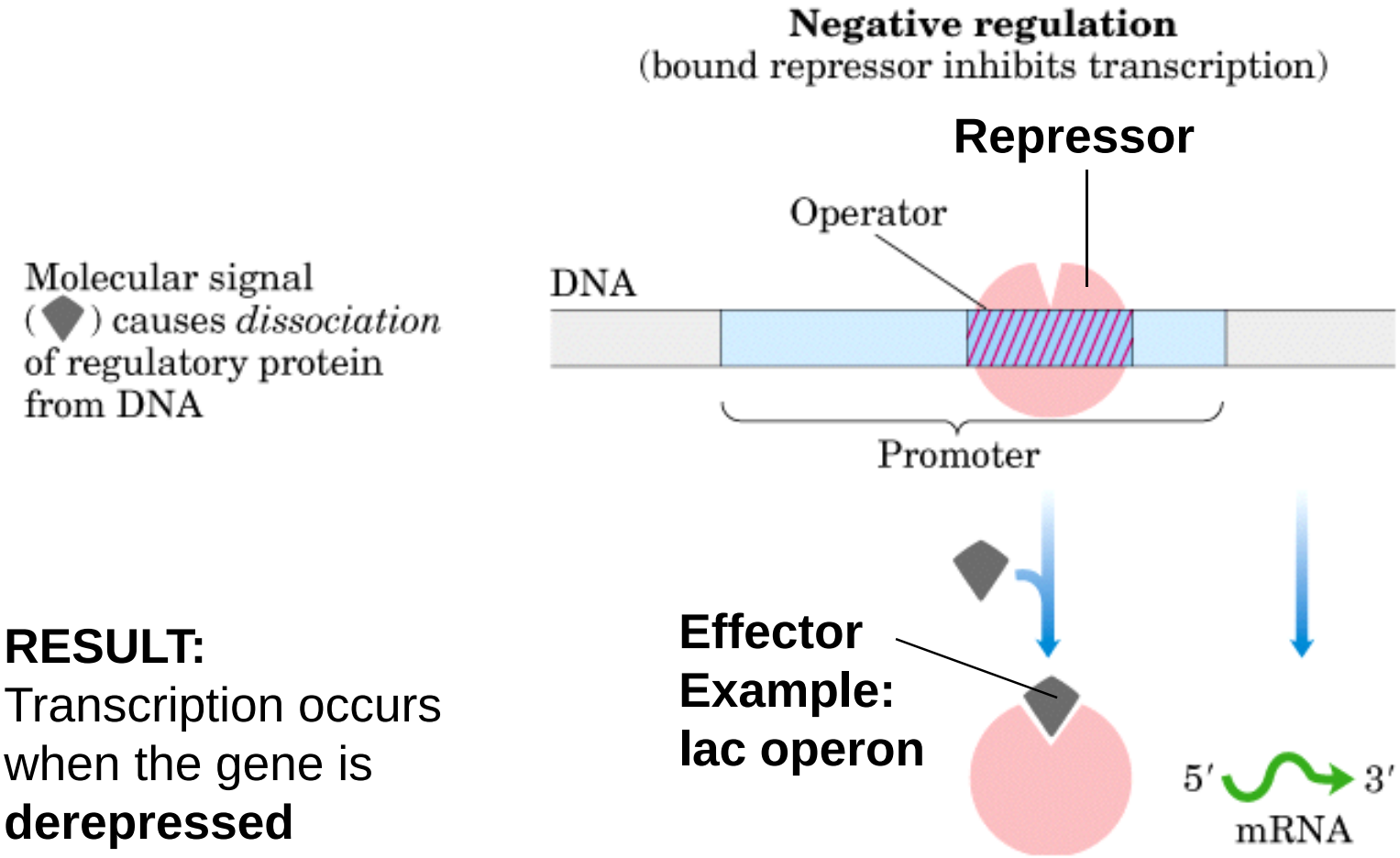
   actively block transcription

   bind to specific "**operator**" sequences (repressor binding sites)

   Repressor binding is modulated by specific **effectors**



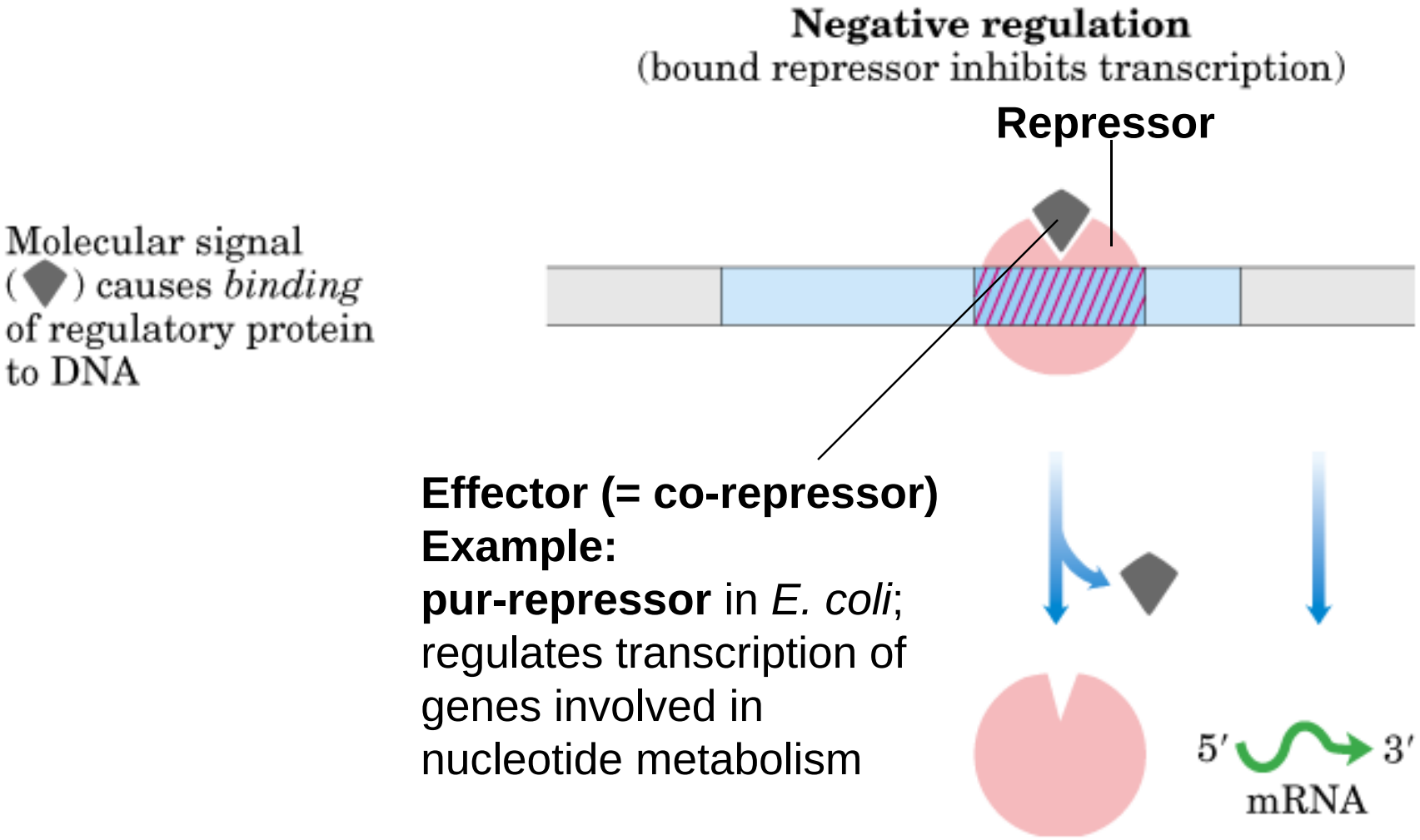
Negative regulation (1)



RESULT:
Transcription occurs when the gene is **derepressed**

Source: Lehninger pg. 1076

Negative regulation (2)



Effector (= co-repressor)
Example:
pur-repressor in *E. coli*; regulates transcription of genes involved in nucleotide metabolism

Source: Lehninger pg. 1076

Modulators of transcription

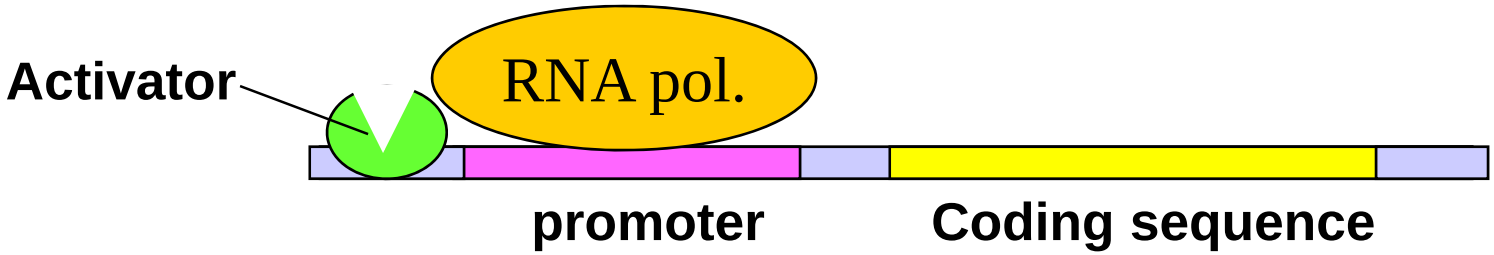
3. Activators:

mediate **positive gene regulation**

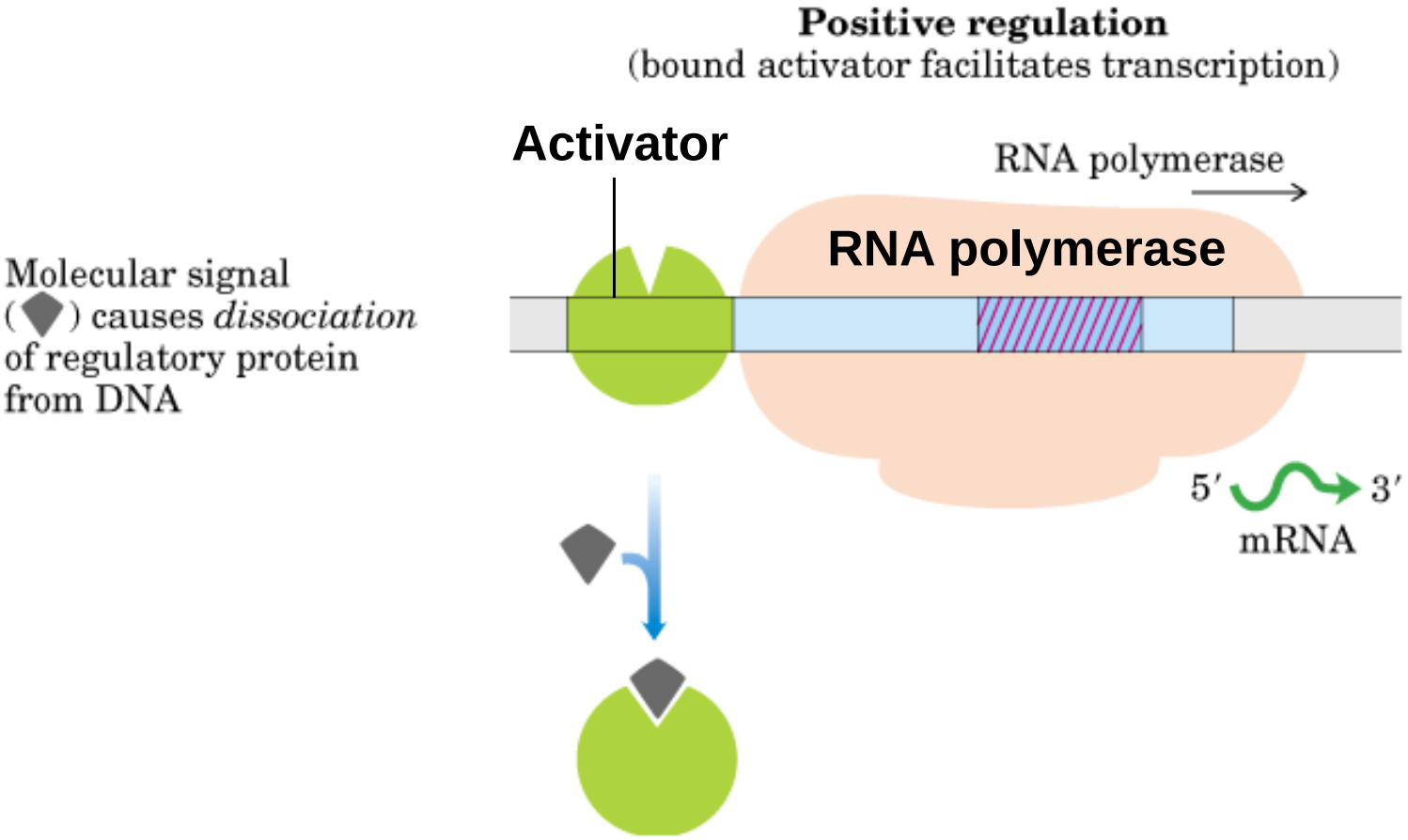
bind to specific regulatory DNA sequences (e.g. **enhancers**)

enhance the RNA polymerase -promoter interaction and actively stimulate transcription

common in eukaryotes

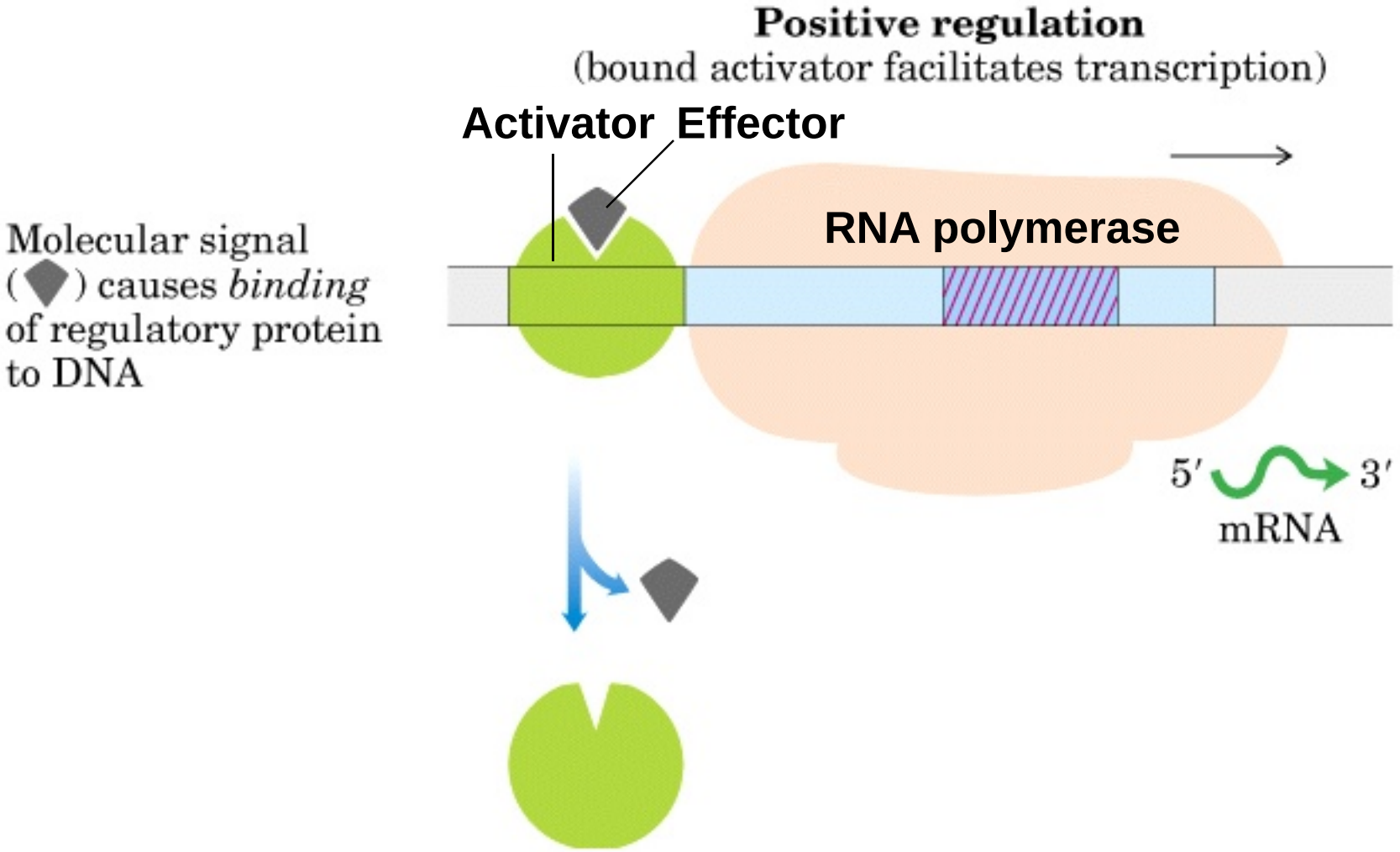


Positive regulation (1)



Source: Lehninger pg. 1076

Positive regulation (2)



Source: Lehninger pg. 1076

Operons

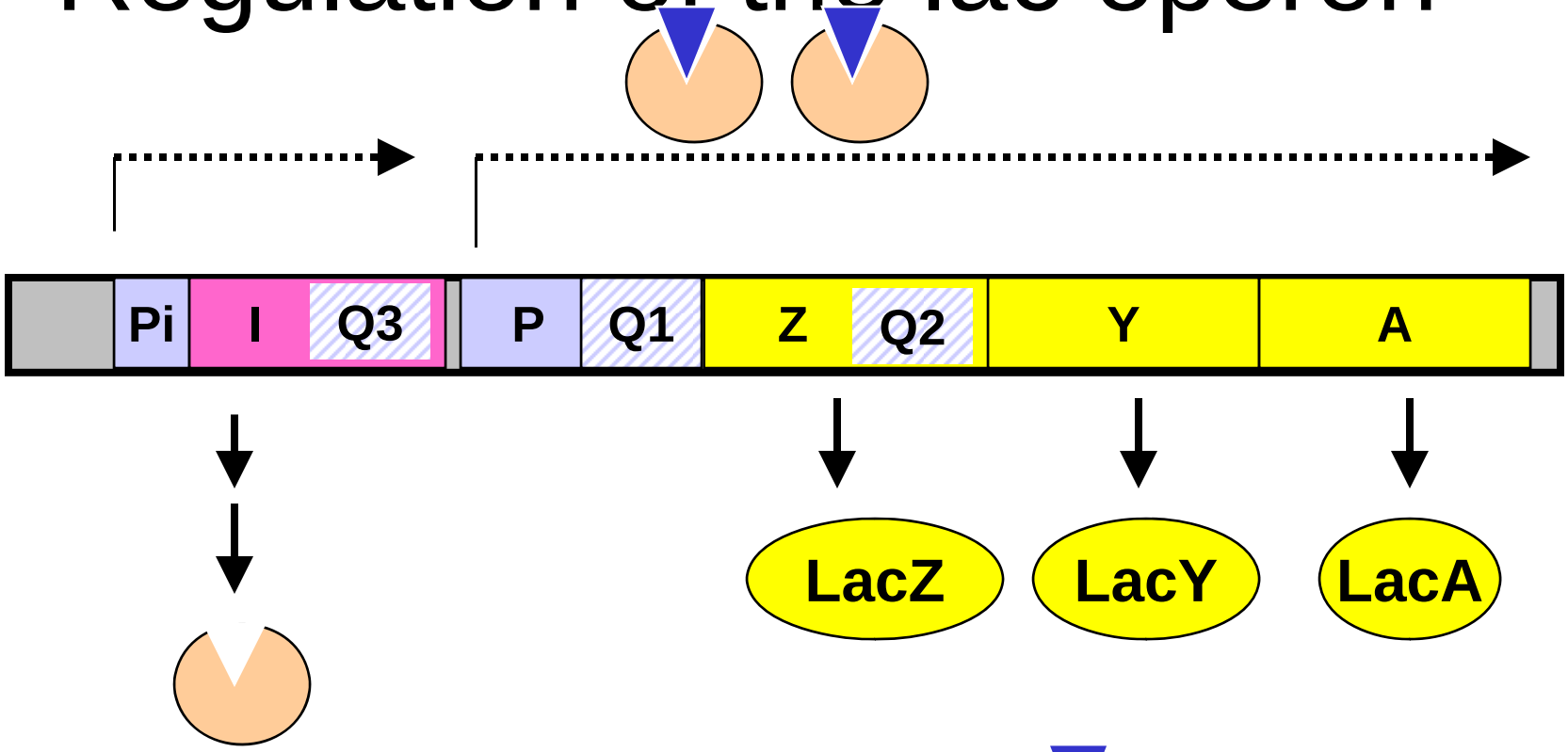
- a promoter plus a set of adjacent genes whose gene products function together.
- usually contain 2 –6 genes, (up to 20 genes)
- these genes are transcribed as a **polycistronic** transcript.
- relatively common in prokaryotes
- rare in eukaryotes

The lactose (lac) operon

- **Contains several elements**
  - *lac*Z gene = β-galactosidase
  - *lac*Y gene = galactosidase permease
  - *lac*A gene = thiogalactoside transacetylase
  - *lac*I gene = *lac* repressor

  - P_i = promoter for the *lac*I gene
  - P = promoter for *lac*-operon
  - O_1 = main operator
  - O_2 and O_3 = secondary operator sites (pseudo-operators)

Regulation of the lac operon



lacI repressor

Inducer molecules:

Allolactose:

- natural inducer, degradable

IPTG (Isopropylthiogalactoside)

- synthetic inducer, not metabolized,

Selected DNA binding motifs

1. **Helix-turn-helix**
   - Homeodomain
2. **Zinc Fingers**
   - Cys4 zinc finger
   - Cys2 His2 zinc finger (e.g. TFIIIA)
3. **Basic domains**
   - Leucine zippers factors (bZIP)
   - Basic helix-loop-helix (bHLH)
4. **Beta-scaffold factors with minor groove contacts**
   - HMG (High mobility group) proteins

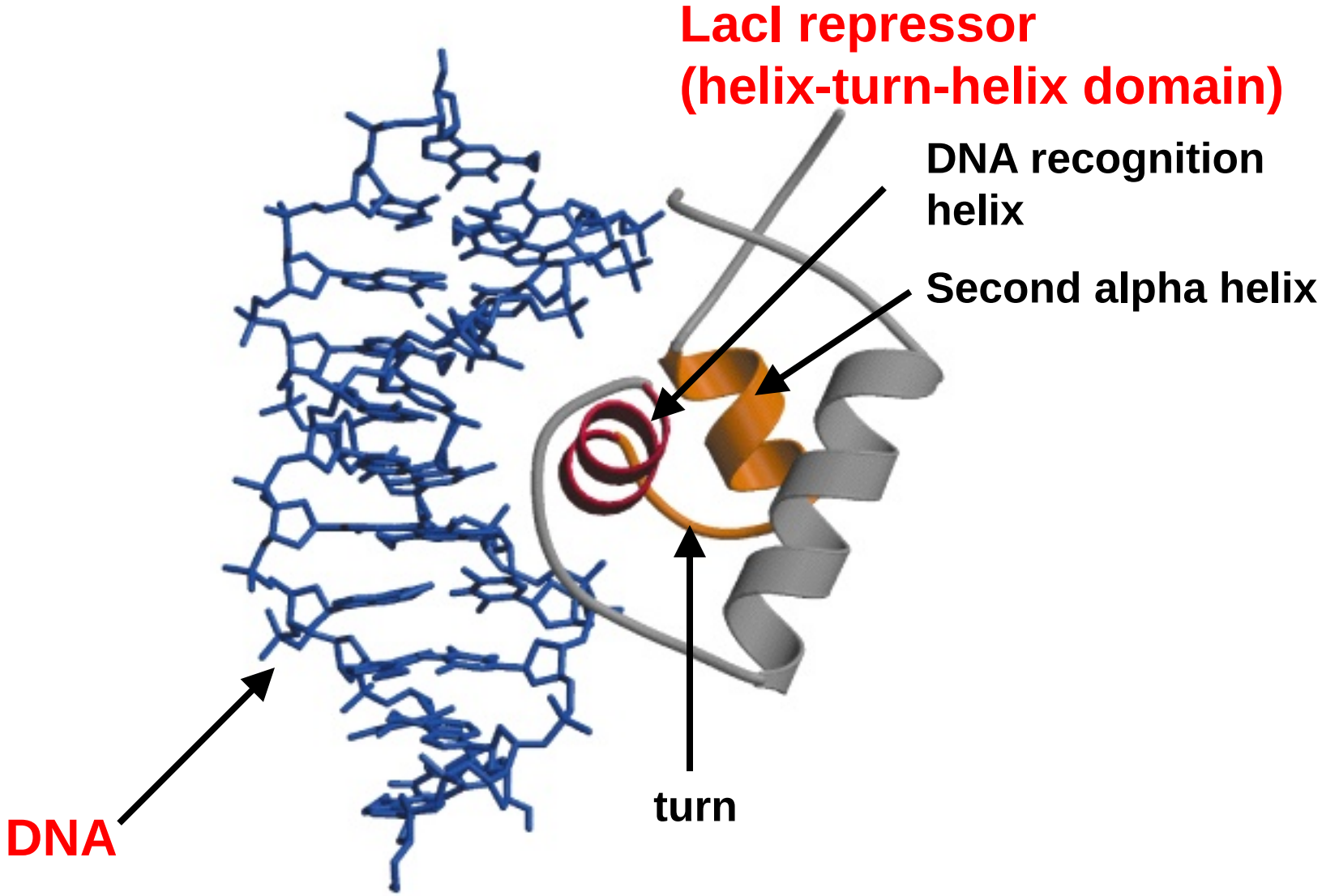
Helix-turn-helix motifs

Structure:

- about 20 amino acids long
- 2 short alpha helicies (7 – 9 amino acids long)
- DNA recognition helix (binds specific DNA sequence)
- Recognition helix and 2nd helix form ~ 90° angle
- very short turn (NOT a beta-turn)
- Often glycine at start of the turn (helix breaker)

GENE	alpha - helix										turn						alpha - helix					
GalR	A	T	I	K	D	V	A	R	L	A	G	V	S	V	A	T	V	S	R	V	I	N
λ-cro	F	G	Q	T	K	T	A	K	D	L	G	V	Y	Q	S	A	I	N	K	A	I	H
P22-cro	G	T	Q	R	A	V	A	K	A	L	G	I	S	D	A	A	V	S	Q	W	K	E
Position			1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20

How does the lac repressor bind DNA?

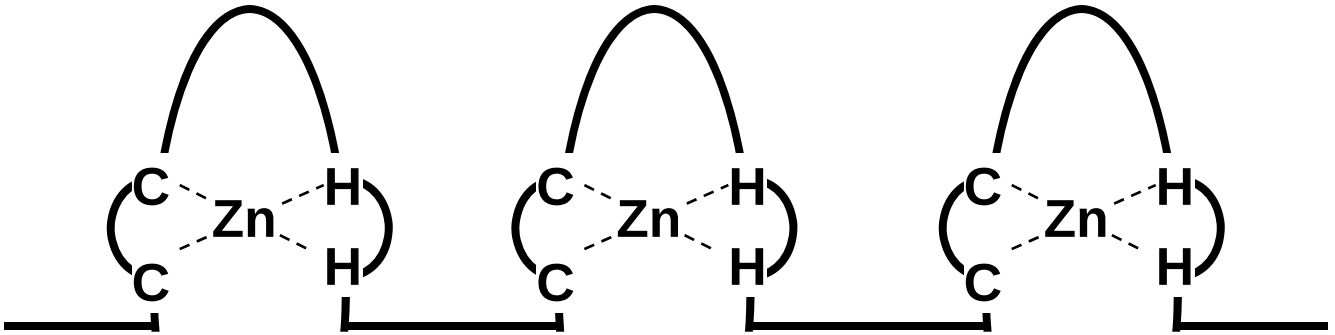


Source: Lehninger pg. 1082

Zinc-Finger Motifs

Several subtypes (Cys4, Cys2-His2 …)

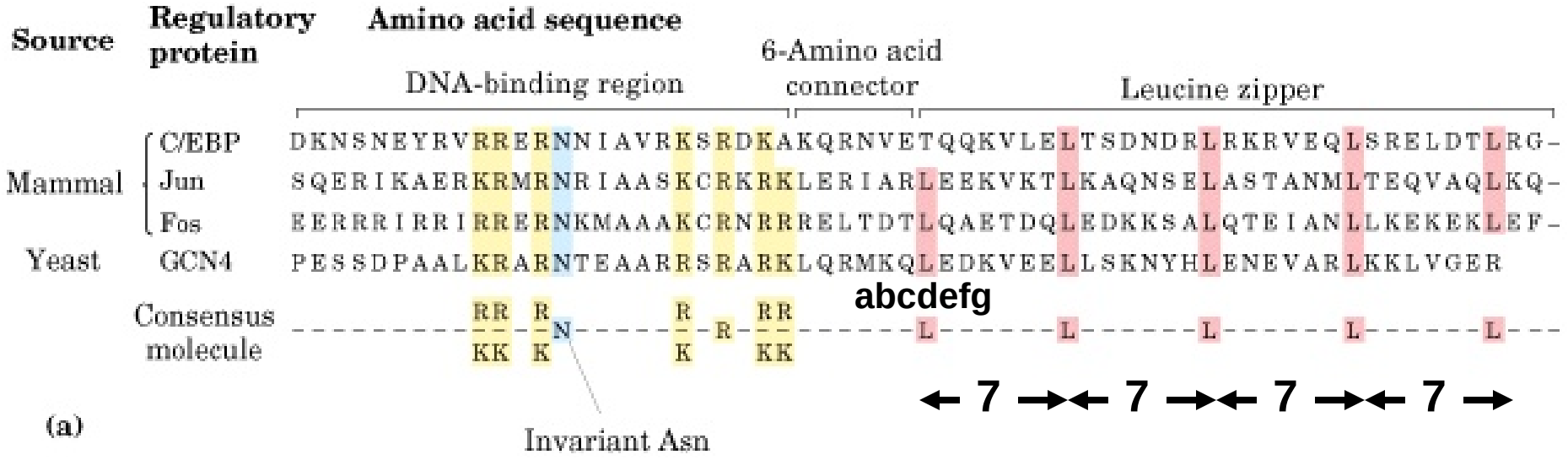
- Example: Cys2 His2 type
- Zinc does not interact with DNA
- Usually multiple zinc-fingers in a row
- At least some also bind RNA
- Consensus sequence:
  [Y,F]-X-C-X_{2-4}-C-XXX-F-XXXXX-L-XX-H-X_{3-5}-H



Basic domains

Leucine zippers (bZip):

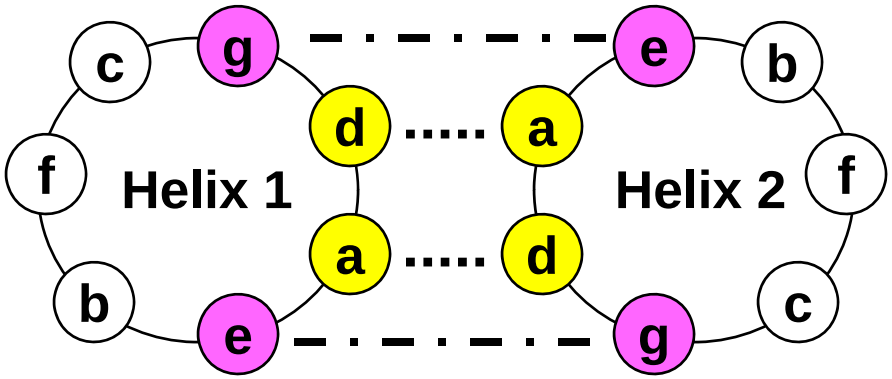
- Basic region of the protein binds to DNA
- Mainly act as dimers or other sometimes as other multimers
- Special alpha-helices allow formation of **coiled-coil** structures.
- Hydrophobic residues (**Leu**) align on one side of the helix
- Example: Jun and Fos

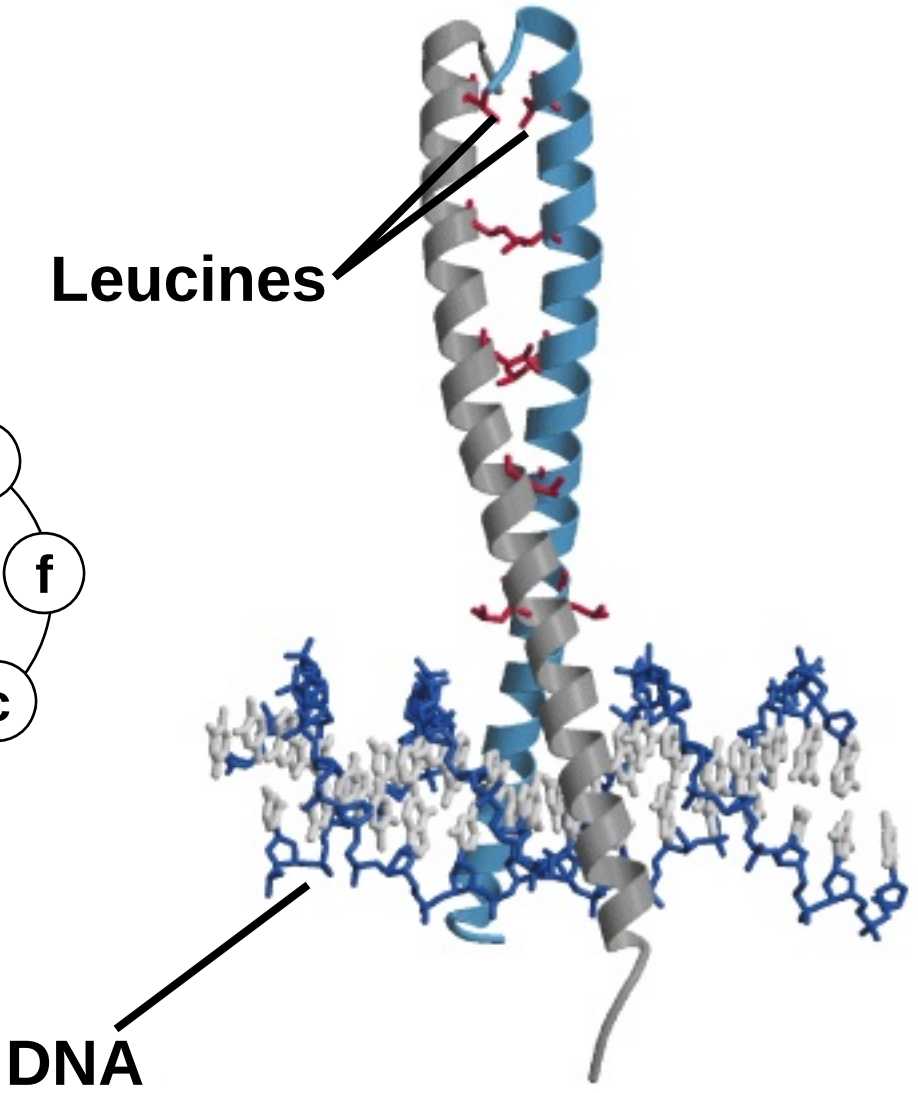


Source: Lehninger pg. 1084

Leucine zippers

3,4 hydrophobic heptad





Source: Lehninger pg. 1084