Regulation of gene expression, molecular basis of signalisation

Molecular Biology

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REGULATION OF GENE EXPRESSION IN PROKARYOTES

Levels of gene expression regulation

REPLICATION

TRANSCRIPTION

↓

POSTTRANSCRIPTION PROCESSING

↓

TRANSLATION

↓

POSTTRANSLATION PROCESSING

Transcription regulation in prokaryotes

- single type of RNA polymerase
- nevertheless genes are take-on and take-off according to needs
- prokaryotic cell must react to quick changes of environmental conditions
- signals for starting and stopping transcription are in the form of small molecules - substrates
- transport of signals to promoters are mediated by protein molecules

Participants on the regulation of gene expression

Regulator

- any substance, which participate on regulation of molecular process

Regulatory protein

- innate entity, which participate on the regulation
- it usually binds to promoter

Allosteric effector

- substance of low molecular mass which by binding to regulatory protein change its conformation and by this its affinity to regulatory region

Types of regulators

Positive regulator

- **induces transcription, translation, …**

Negative regulator

- **inhibite transcription, translation, …**

Allosteric effect

= interaction of an effector with regulatory protein

↓

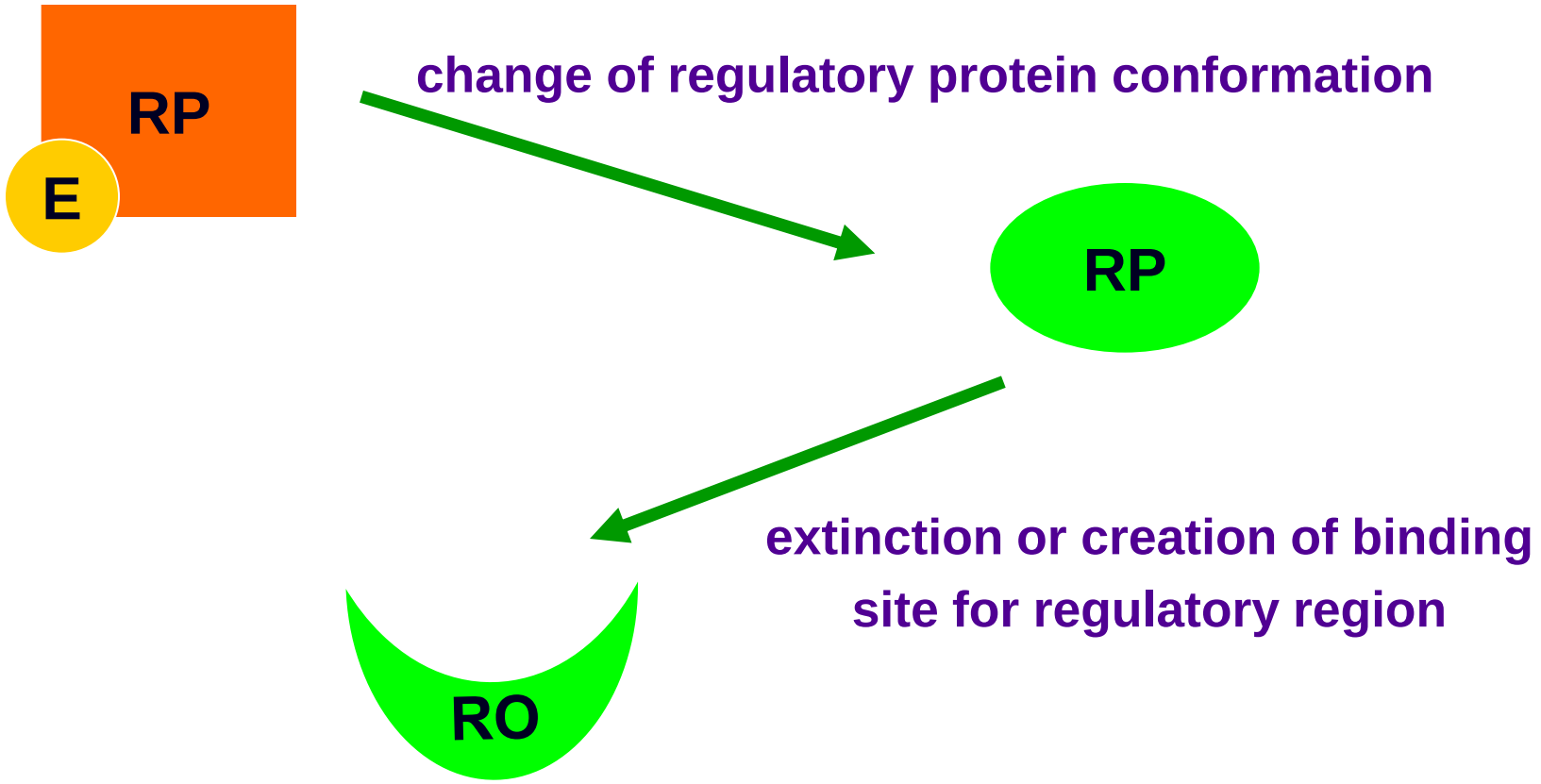
change of regulatory protein conformation

↓

extinction or creation of binding site for regulatory region

Alosteric effect

= interaction of an effector with regulatory protein



Types of allosteric effectors

Negative allosteric effector

= inhibites regulatory protein binding to regulatory region

Positive allosteric effector

= facilitates regulatory protein binding to regulatory region

Types of regulatory proteins

Negative regulatory proteins

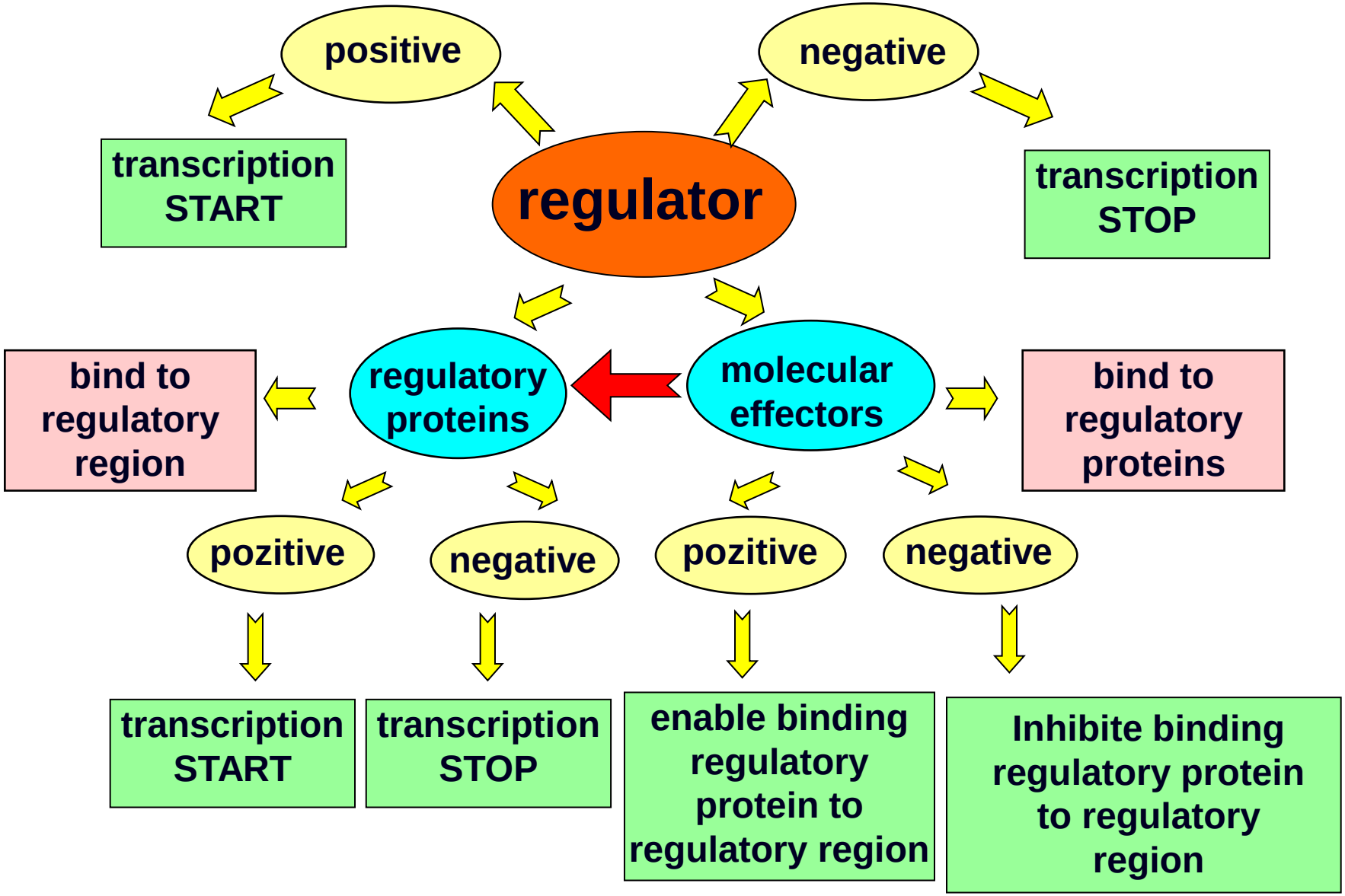
= their binding to regulatory region inhibite transcription of transcription unit by RNA polymerase

Positive regulatory proteins

= their binding to regulatory region enable transcription of transcription unit by RNA polymerase

= activator of transcription

Net of regulatory internships



Negative and positive regulation of operon

NEGATIVE

- it is a keystone of enzymatic induction and repression
- binding of active repressor to operator stops transcription

POZITIVE

- it is a keystone of catabolic repression
- binding of CAP to promoter in the presence of inductor stimulates transcription

Enzymatic induction

- **Mostly for synthesis of enzymes which is stimulated by inducer**

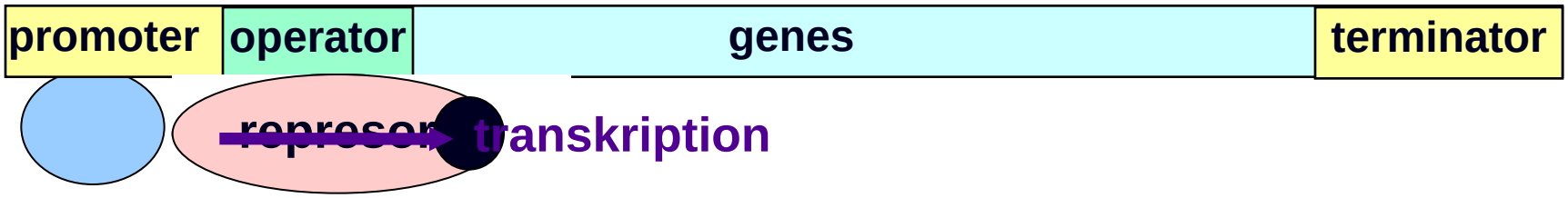
Inducible enzymes

Constitutive enzymes

- **their synthesis do not depend on the presence of inductor**
- **they are produced in cell in constant amount**

Enzymatic induction

induktor



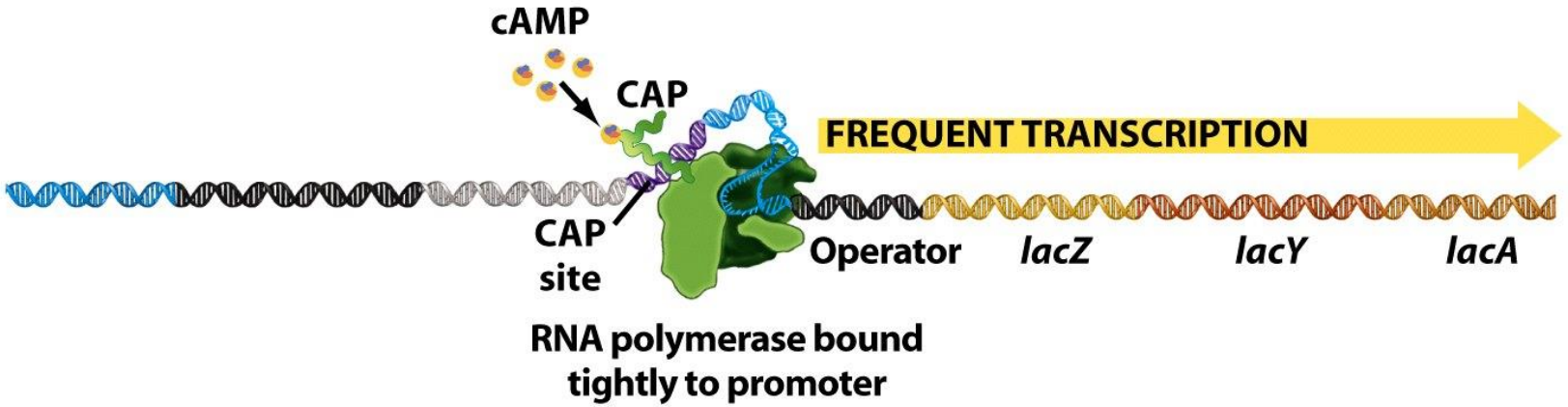
inducer – repressor inactivation

inactive complex

transcription of genes is ON – inducible enzymes are produced

Enzymatic induction

(a) When cAMP is present, it binds to CAP. The cAMP-CAP complex binds to DNA at the CAP site and increases binding of RNA polymerase to promoter. Transcription occurs frequently.



(b) When cAMP is absent, CAP does not bind to DNA. RNA polymerase does not bind the promoter efficiently, and transcription occurs rarely.

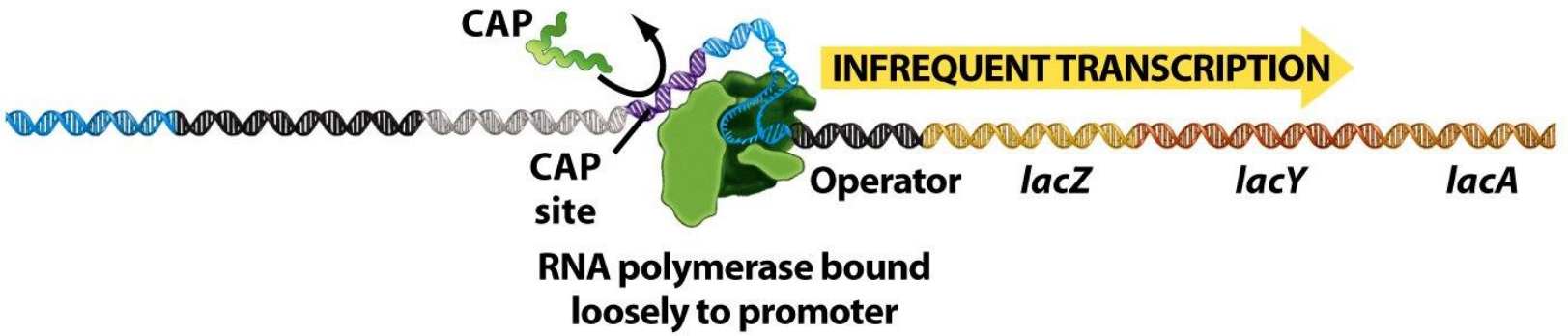


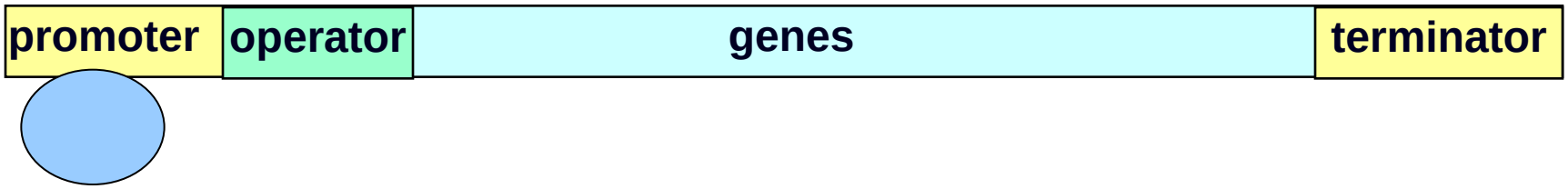
Figure 17-8 Biological Science, 2/e

Enzymatic repression

- **Mostly for the enzymes of biosynthetic pathways**
- **The synthesis of these enzymes is inhibited by a specific metabolite of suitable metabolic pathway which is accumulated to critical amount and stops next synthesis**
- **The synthesis is renewed if concentration of the metabolite fall down under the critical amount**

Enzymatic repression

represor

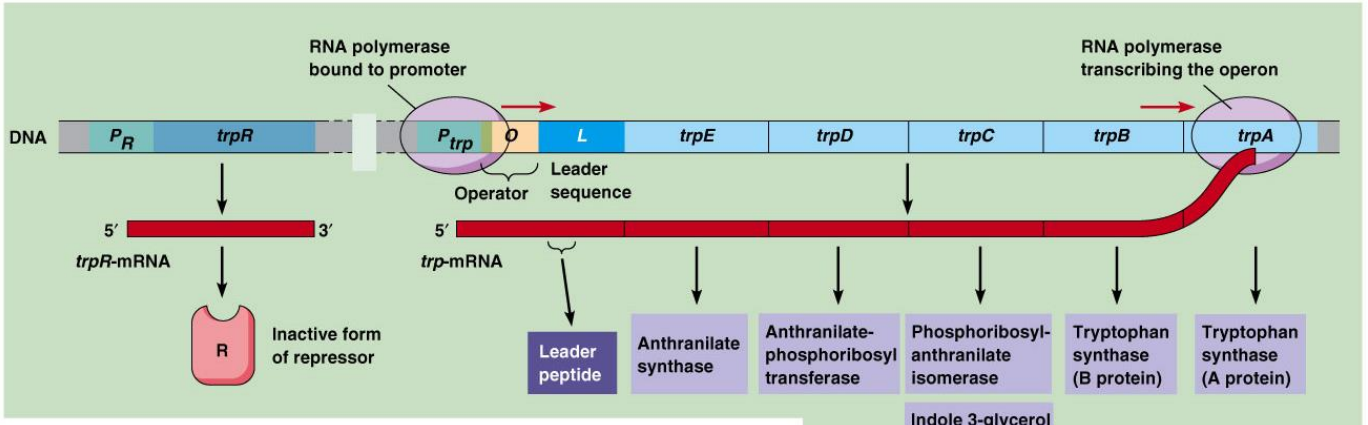


corepressor – activates repressor

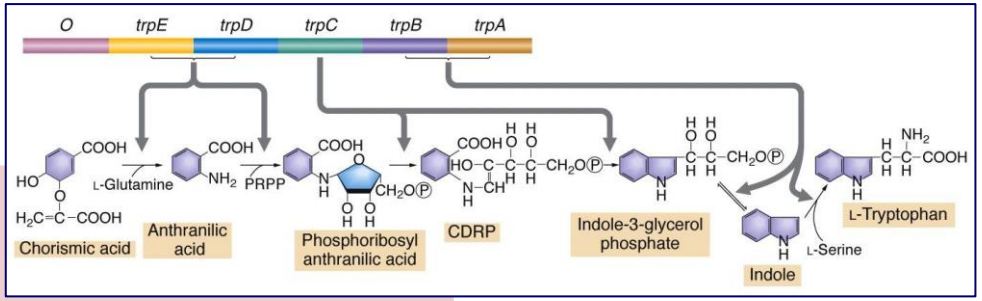
repressor

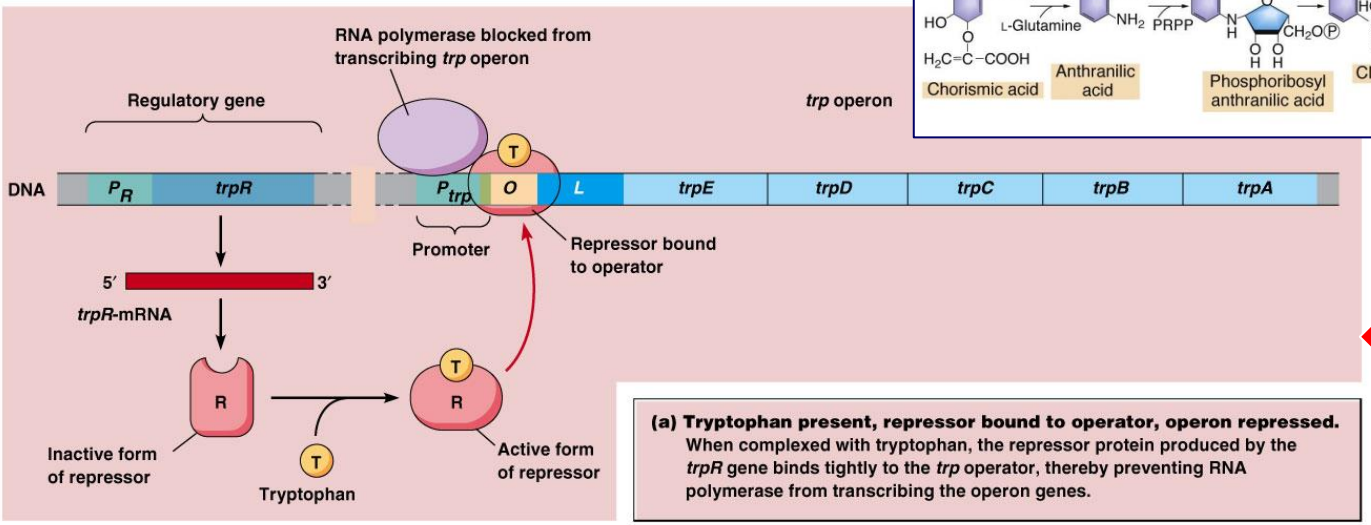
transcription of gene is OFF – synthesis of corepressor is stopped

Enzymatic repression



(b) Tryptophan absent, repressor not bound to operator, operon derepressed. In the absence of tryptophan, the free *trp* repressor cannot bind to the operator site. RNA polymerase can therefore move past the operator and transcribe the *trp* operon genes, giving the cell the capability to synthesize tryptophan.





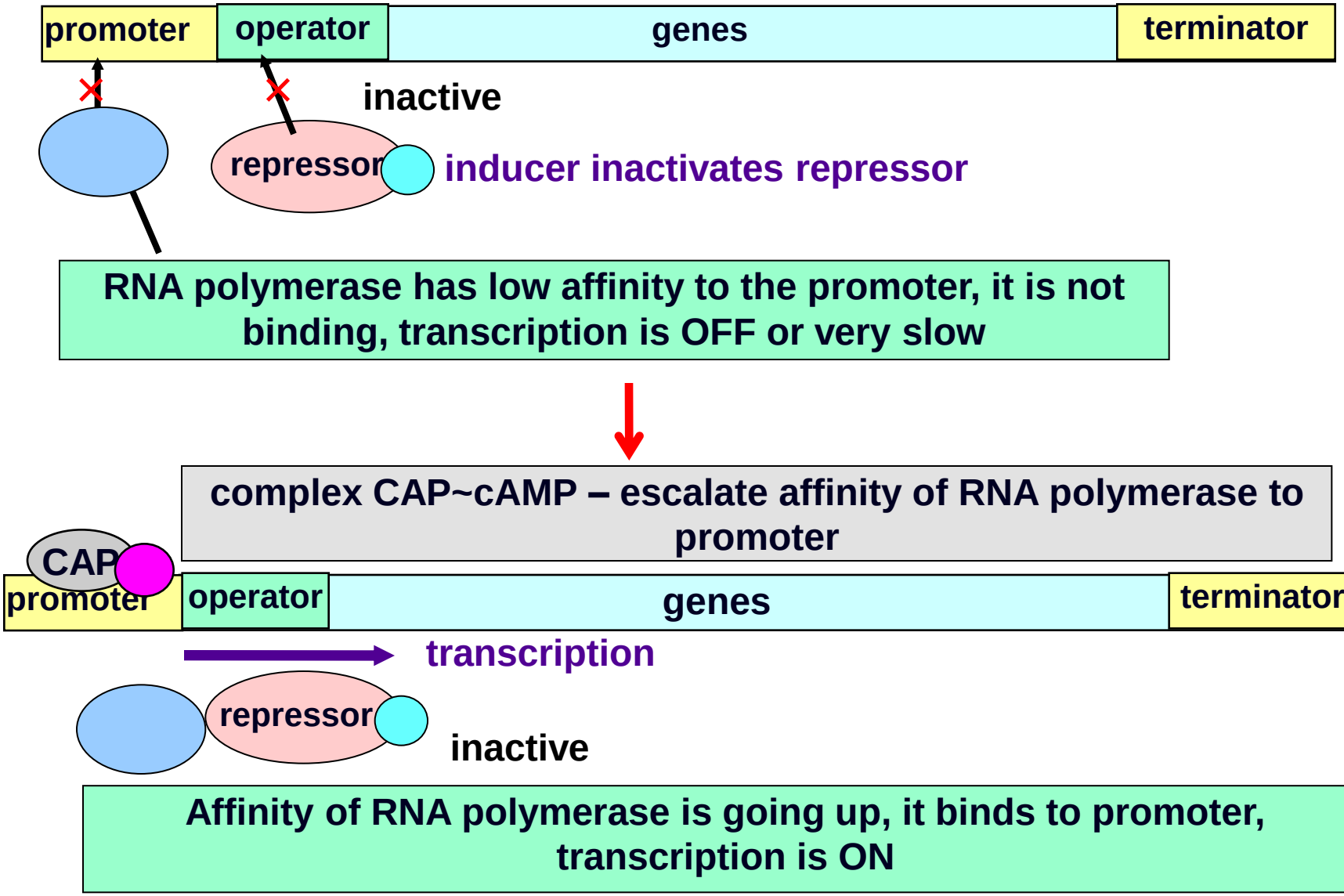
(a) Tryptophan present, repressor bound to operator, operon repressed. When complexed with tryptophan, the repressor protein produced by the *trpR* gene binds tightly to the *trp* operator, thereby preventing RNA polymerase from transcribing the operon genes.

http://www.mun.ca/biology/scarr/bio4241_chapter13.htm

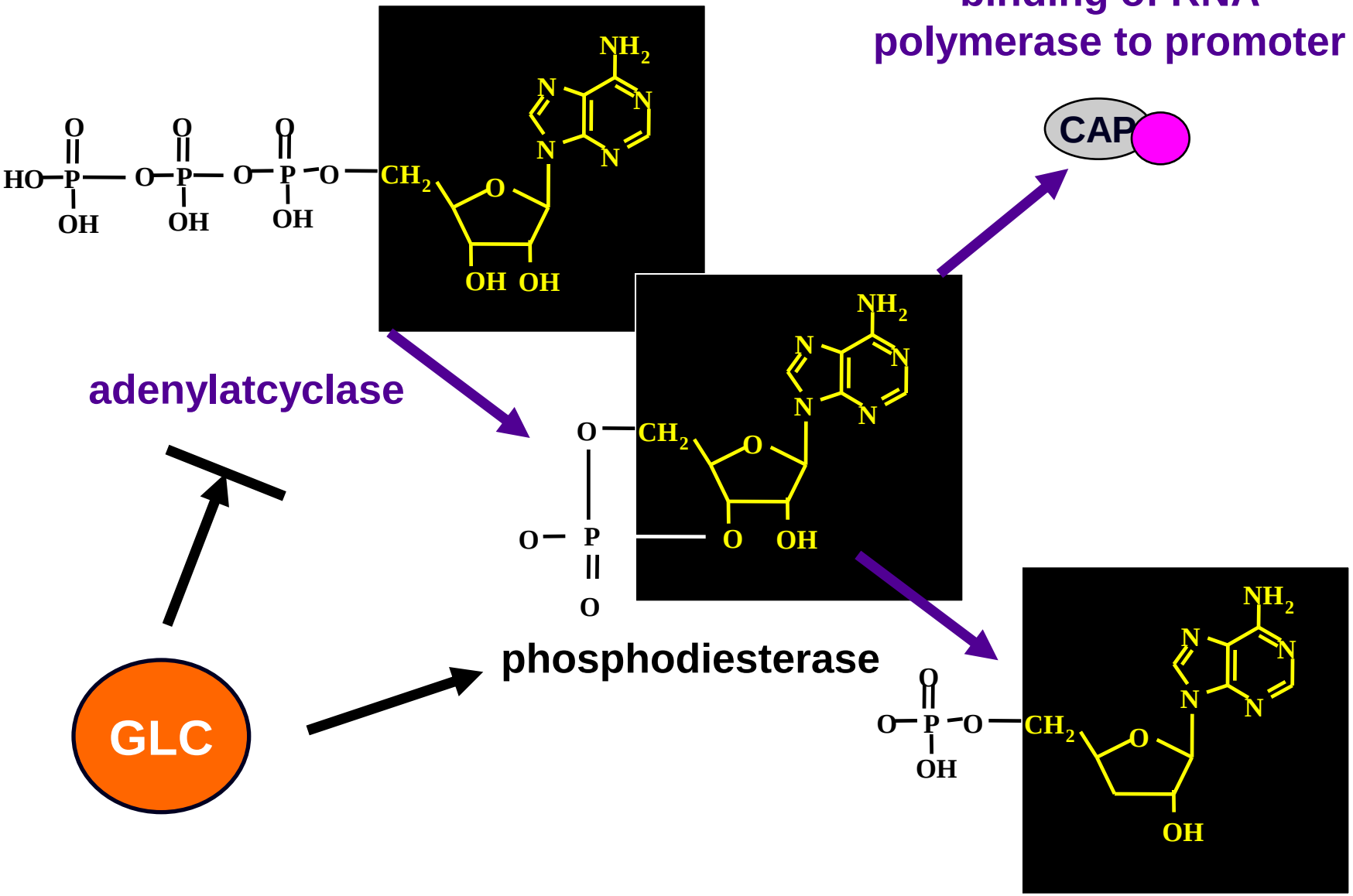
Catabolic repression

- **Substrate inhibits synthesis of inducible enzymes although inducer is still present**
- **For example, glucose inhibits synthesis of β-galactosidase even in the presence of lactose as an inducer**

Catabolic repression

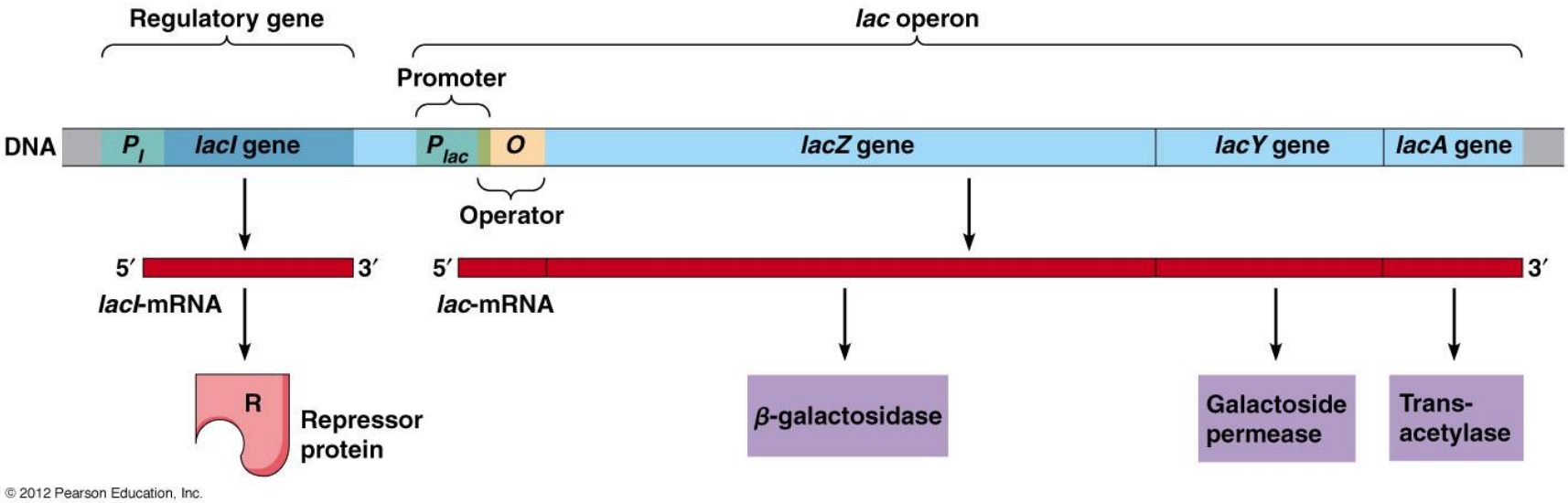


Function of glc in catabolic repression



lac operon of Escherichia coli

the enzymes for metabolism of lactose



I: codes repressor

Z: codes enzyme β-galaktosidase (lac → glu + gal)

Y: codes enzyme permease

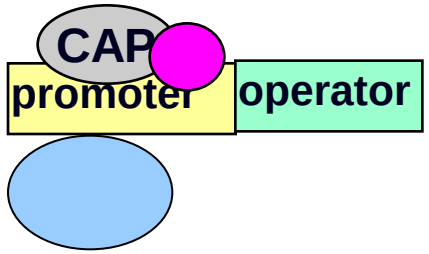
A: codes enzyme thiogalaktosidtransacetylase

Inducer = lactose

Regulation of lac operone

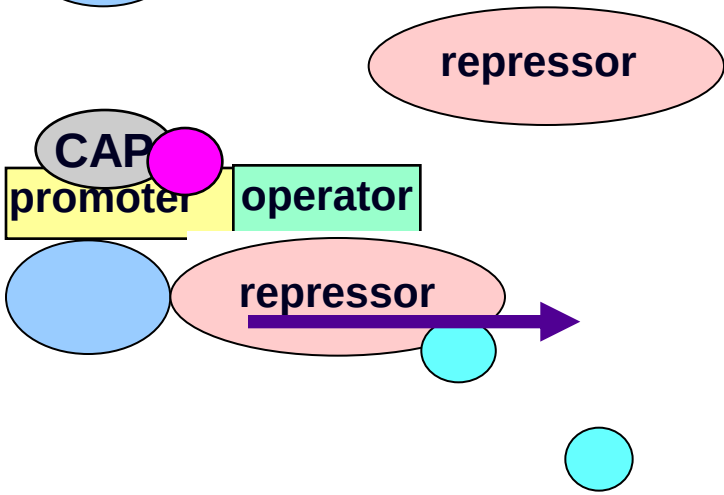
a) glc is not present, lac is not present

→ repressor binds to operator



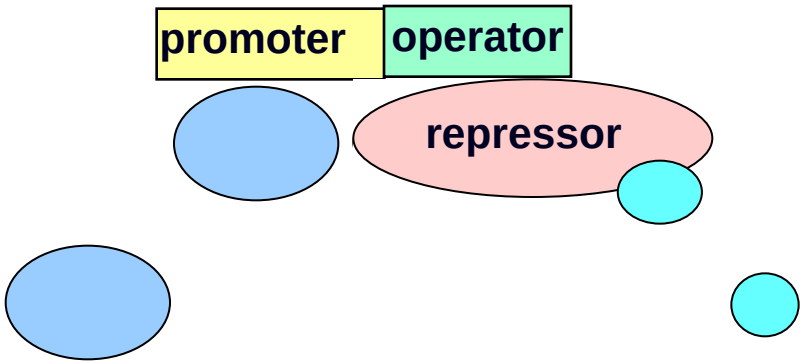
b) glc is not present, lac is present

enzyme induction



c) glc is present, lac is present

catabolic repression



Regulace lac operonu

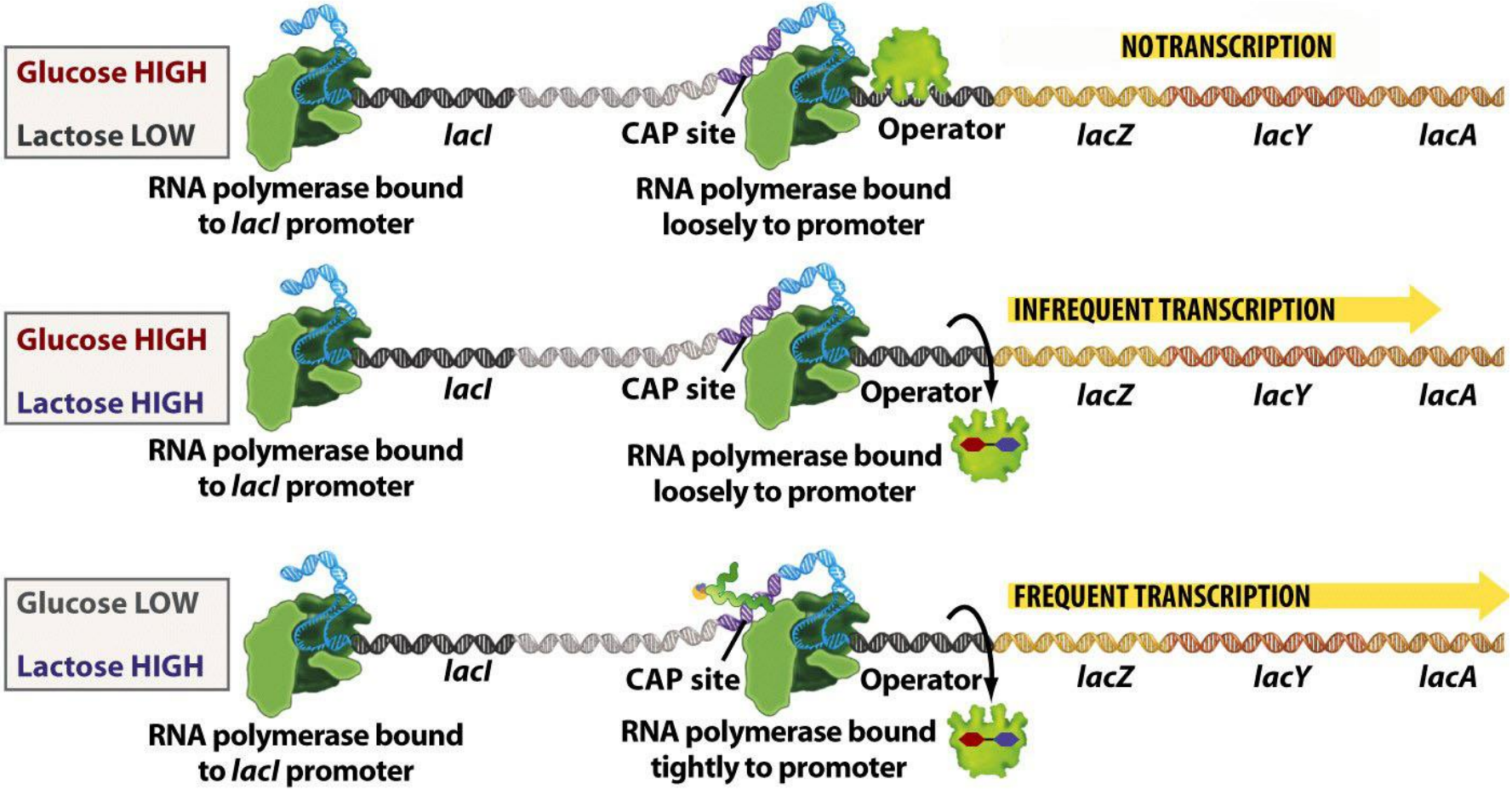


Figure 17-10 Biological Science, 2/e

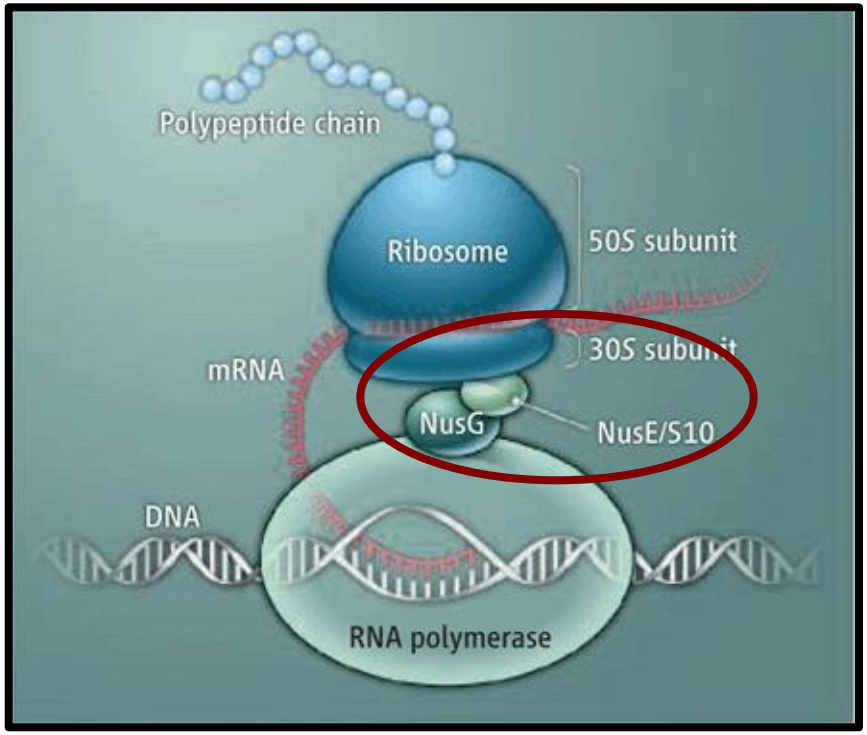
Attenuation

- **The process of regulation of transcription based on the attenuator**
- **Attenuator is a part of leader sequence of gene**
- **Attenuator acts as an early terminator of transcription**

for example *trp* operon

contains genes necessary for tryptofan synthesis

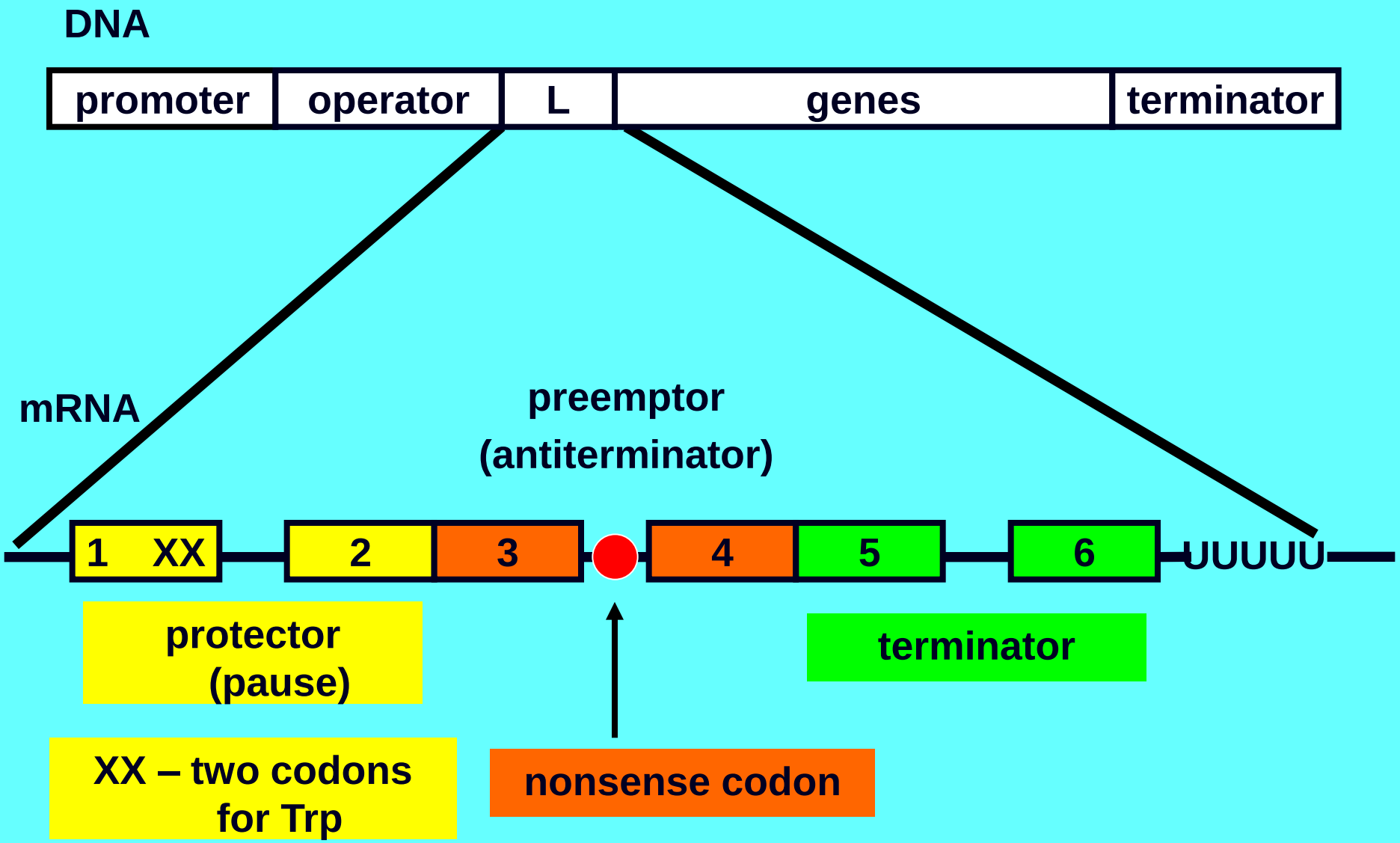
Do you still remember ?



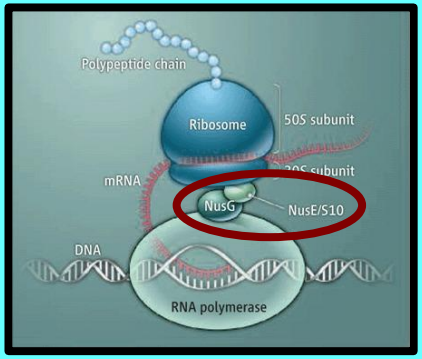
J. W. Roberts Science 328, 436-437 (2010)

Science
AAAS

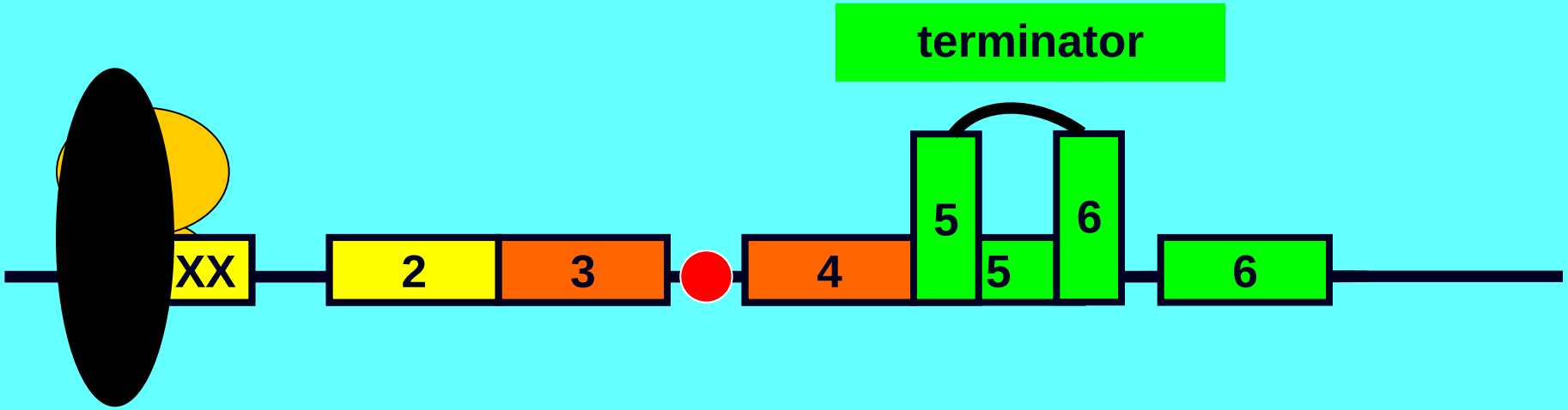
Structure of attenuator



Attenuation in redundancy of Trp



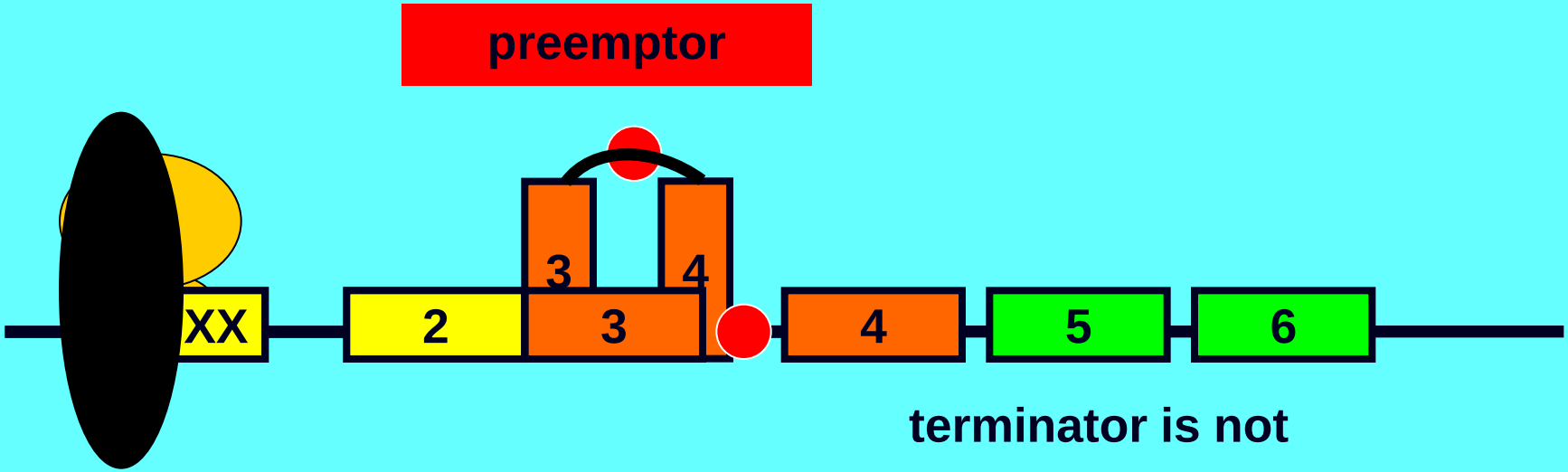
RNA polymerase releases here



translation stops on the nonsense codon

TRANSCRIPTION IS STOPPED

Attenuation in deficiency of Trp



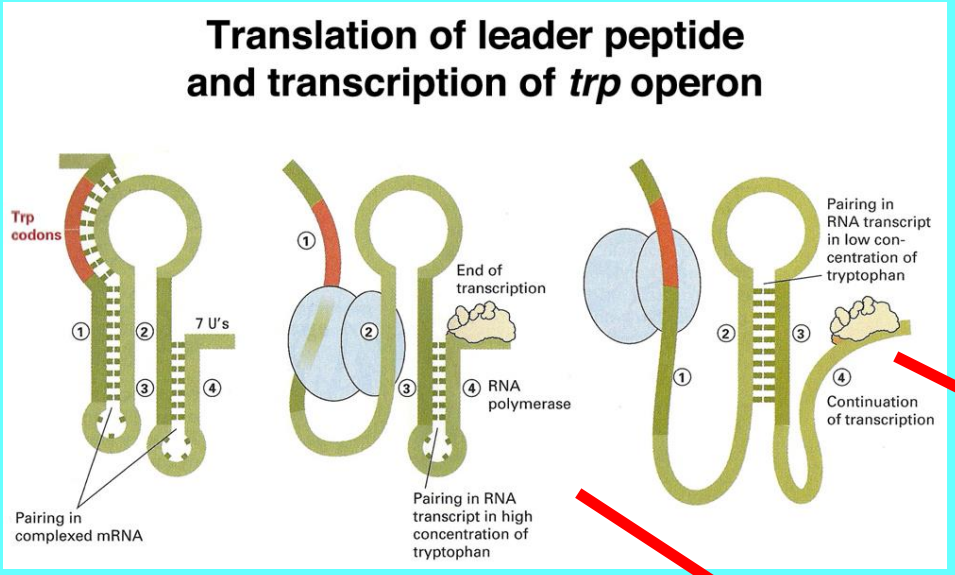
terminator is not formed

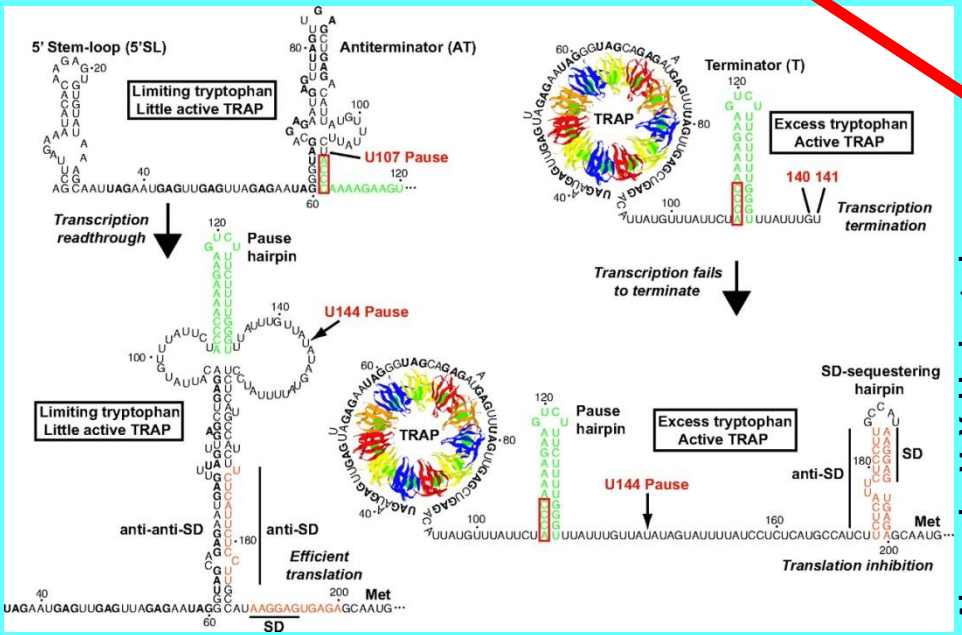
Translation stops on the codons for Trp because this amino acid is in low number of copies

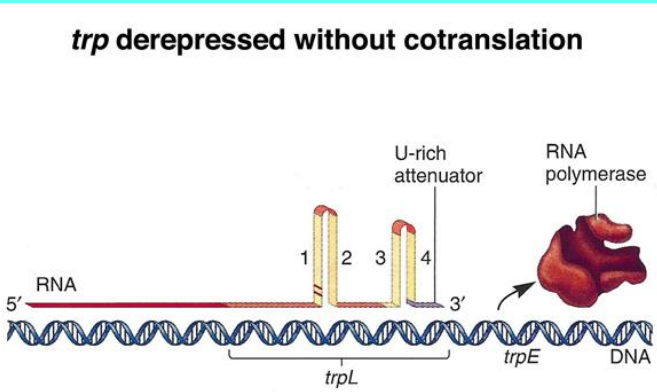
TRANSCRIPTION FOLLOWS

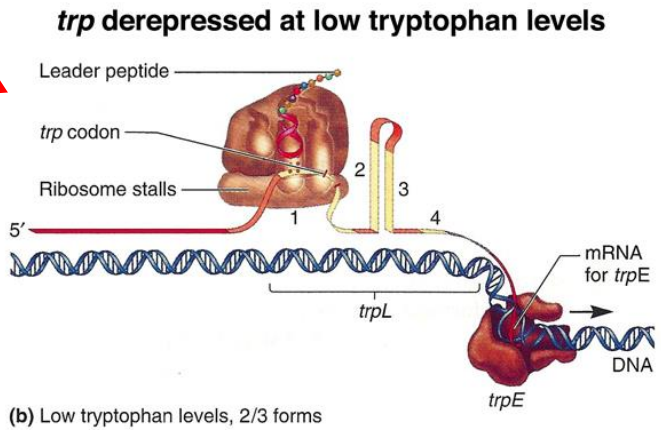
https://www.youtube.com/watch?v=CeE83RyQFRo

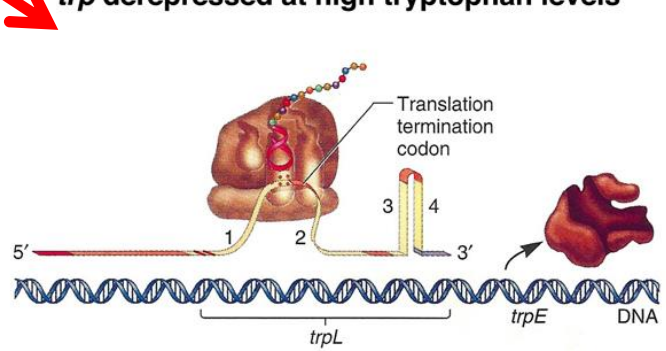
Trp attenuation









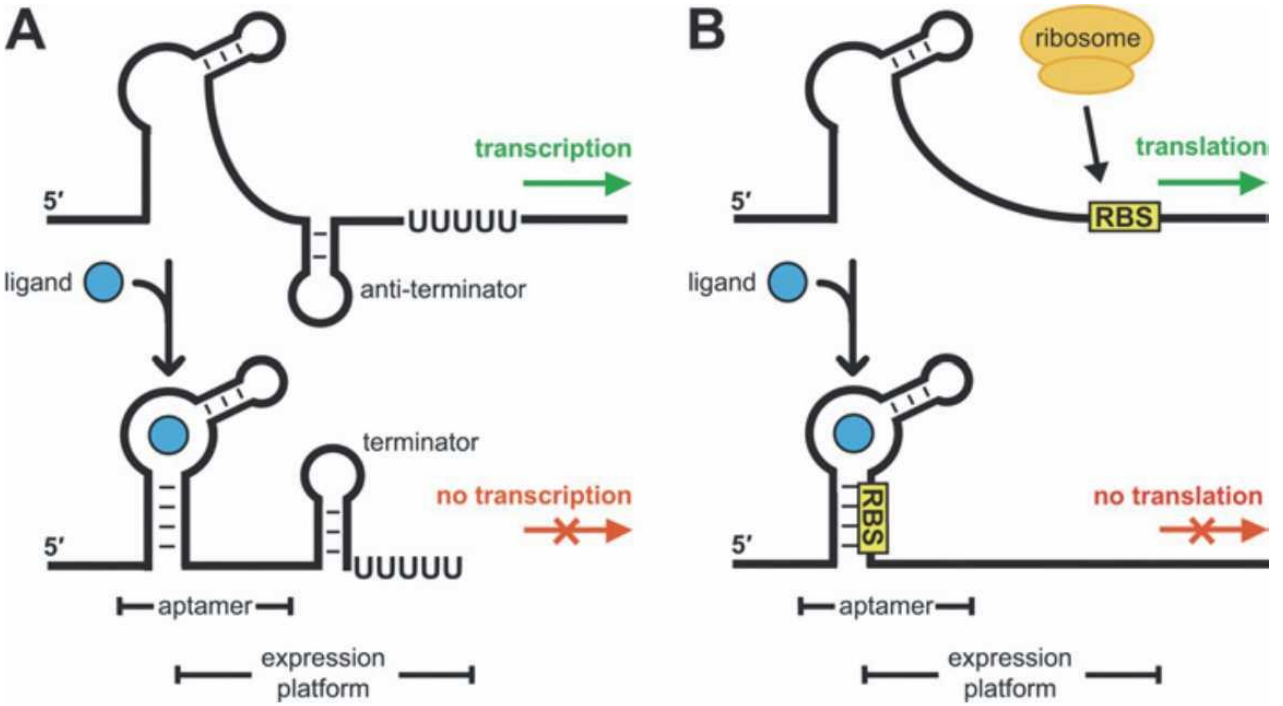


Alexander V. Yakhnin et al.
PNAS 2008;105:16131-16136

http://www.discoveryandinnovation.com/BIOL202/notes/lecture17.html

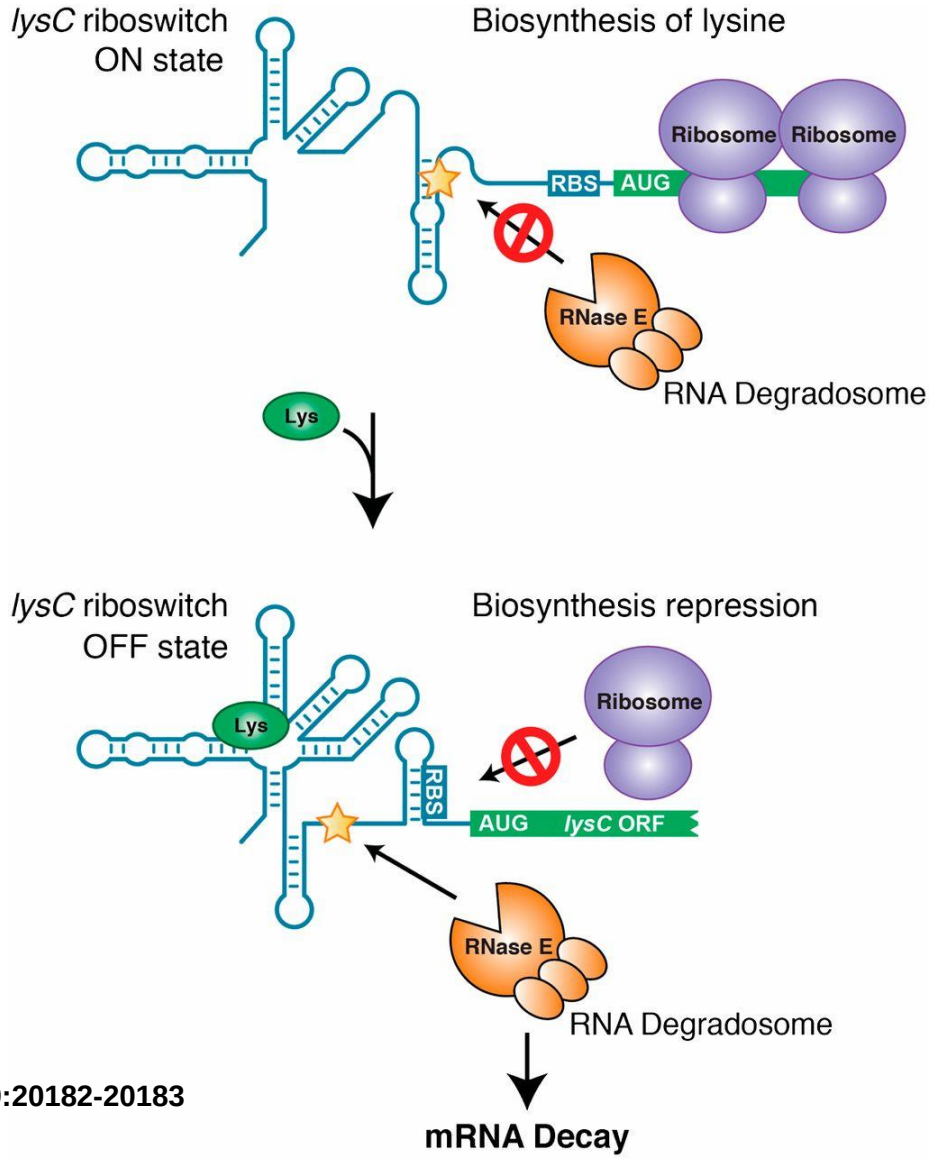
Riboswitches

- **Secondary mRNA structure capable to bind molecular effector (ligand)**
- **Binding of ligand on mRNA changes its secondary structure ⇒ the gene expression is „ON“ or „OFF“**



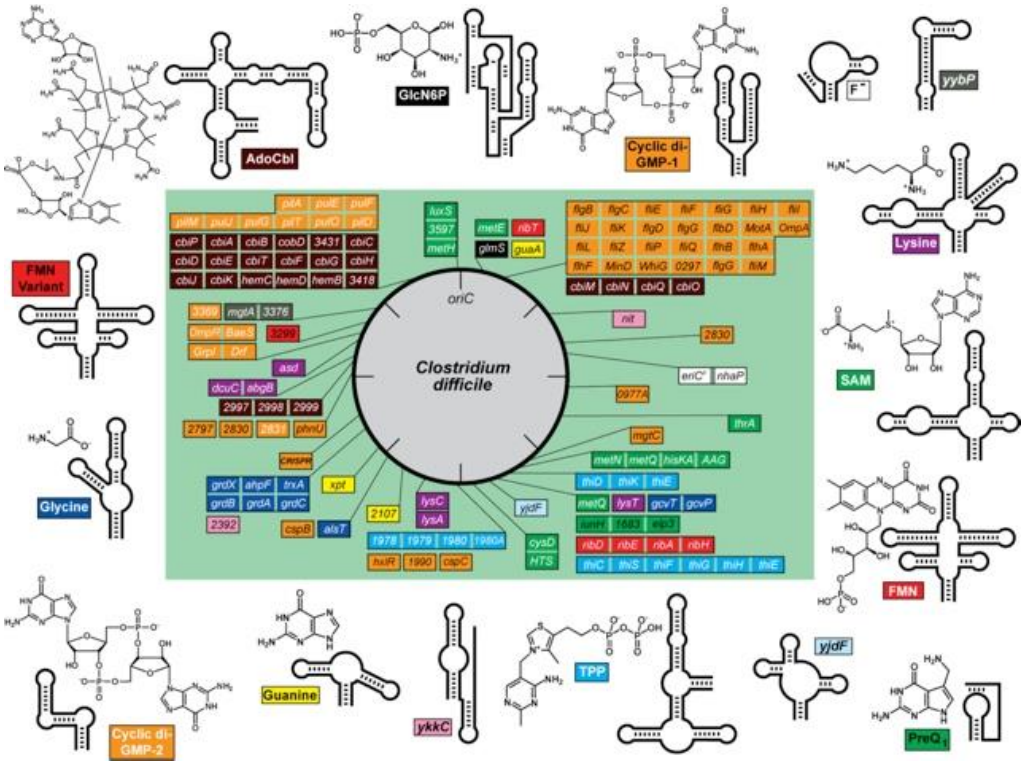
https://sites.lsa.umich.edu/walter-lab/research/riboswitch/

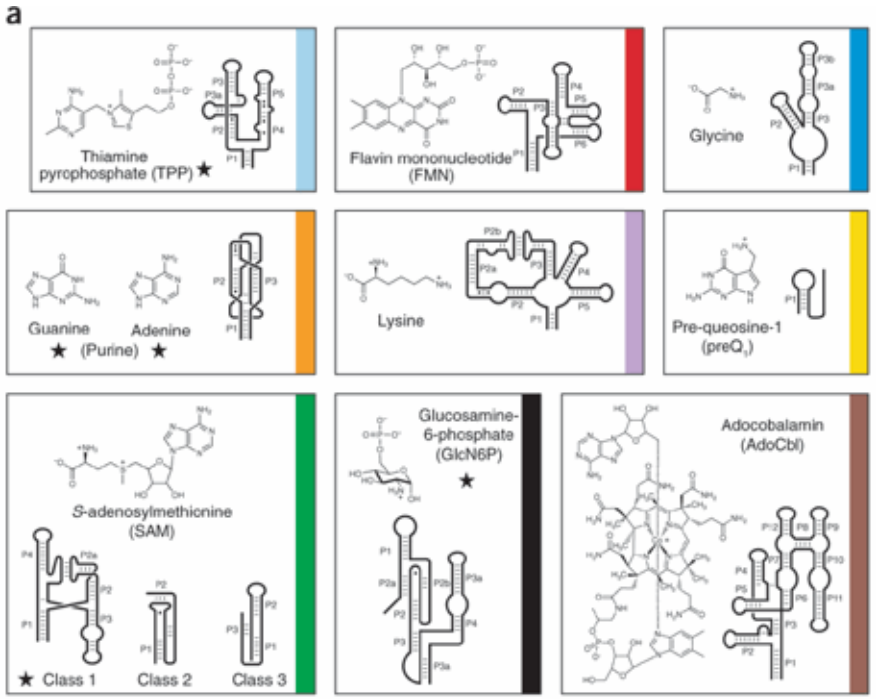
Regulatory mechanisms of the lysC riboswitch controlling translation initiation and mRNA decay



Marie-Pier Caron et al. PNAS 2012;109:20182-20183

Other types of riboswitches





b

Riboswitch target metabolite:	TPP	FMN	AdoCbl	Purine	SAM1	SAM2	SAM3	Lysine	GlcN6P	Glycine	PreQ1
Acinetobacter baumannii	1(3)	1(1)	1(1)	–	–	–	–	–	–	1*(1)	–
Bacillus anthracis	6(19)	2(5)	1(1)	6(9)	17(36)	–	–	4(4)	1(2)	1(1)	2(5)
Brucella melitensis	2(11)	1(1)	2(5)	–	–	1(1)	–	–	–	1*(3)	–
Enterococcus faecalis	2(5)	–	2(4)	1(2)	–	–	1(1)	1(1)	1(1)	–	2(3)
Escherichia coli	3(11)	1(1)	1(2)	–	–	–	–	1(1)	–	–	–
Francisella tularensis	1(1)	1(5)	–	–	–	1(1)	–	–	–	–	1(1)
Haemophilus influenza	3(11)	1(1)	–	–	–	–	–	1(1)	–	1*(1)	1(1)
Helicobacter pylori	1(2)	–	–	–	–	–	–	–	–	–	–
Listeria monocytogenes	2(5)	1(1)	2(20)	2(3)	7(14)	–	–	1(1)	1(1)	1(3)	1(1)
Mycobacterium tuberculosis	2(6)	–	2(4)	–	–	–	–	–	–	2*(3)	–
Pseudomonas aeruginosa	1(1)	1(2)	5(24)	–	–	–	–	–	–	–	–
Salmonella enterica	3(11)	1(1)	2(22)	–	–	–	–	–	–	–	–
Staphylococcus aureus	2(7)	2(5)	–	1(2)	4(6)	–	–	2(2)	1(1)	1(3)	2(4)
Streptococcus pneumoniae	4(11)	1(4)	–	1(2)	–	–	1(1)	–	–	1(1)	–
Vibrio cholerae	2(7)	1(1)	1(1)	–	–	–	–	3(3)	–	1(1)	–
Yersinia pestis	3(8)	1(1)	1(2)	–	–	–	–	–	–	–	–

http://www.yalescientific.org/2013/04/a-genetic-light-switch-riboswitches-shed-light-on-the-history-of-life/

Heat shock proteins

- Proteins which are induced by higher temperature
- They are expressed by higher speed in the temperatures 42-45°C
- Regulated by sigma factor sigma-32, which binds to other promoters than the standard sigma factor
- Auto regulation by feedback loop = after accumulation the induced protein binds to sigma factor and releases it from RNA polymerase

Regulation the genes for rRNA and tRNA

- **Speedy of cell division is connected to speedy of ribosome formation**

- **Synthesis of rRNA and tRNA is TAKE OFF if level of amino acid pool is low, synthesis of mRNA continues at least minimum amount**