Regulation
of Bacterial Gene Expression

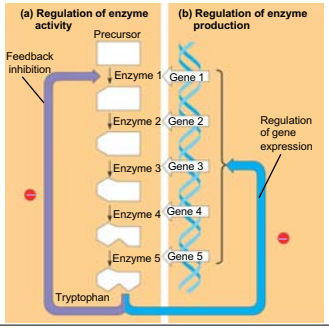


Figure 18.20a, b

Metabolic Regulation

1. Adjust the activity of metabolic enzymes already present
2. Regulate the genes encoding the metabolic enzymes

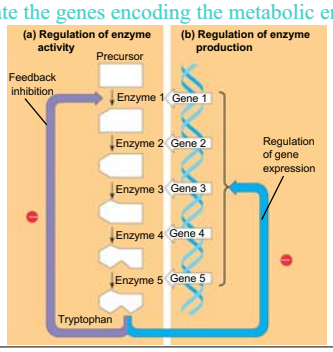
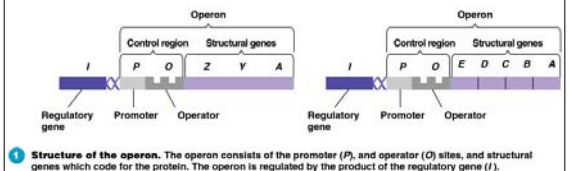


Figure 18.20a, b

Operons: functional gene clusters

- In bacteria, genes are often clustered into **operons**, composed of
  - A **promoter**
    - Site for RNA-polymerase to bind and initiate transcription
  - An **operator**, the "on-off" switch
    - Region of DNA within the promoter or between the promoter and the first gene
  - The **genes** for metabolic enzymes
    - Usually a set of enzymes catalyzing different steps in a common metabolic pathway
    - All the genes in the operon set are transcribed onto a single, common mRNA

Operons: functional gene clusters



Structure of the operon. The operon consists of the promoter (*P*), and operator (*O*) sites, and structural genes which code for the protein. The operon is regulated by the product of the regulatory gene (*I*).

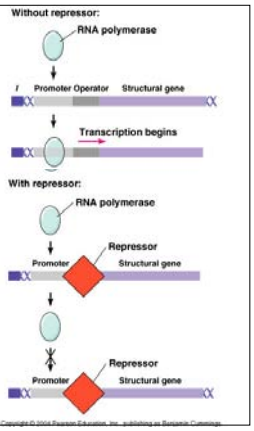
Regulation of Bacterial Gene Expression

- Conserve energy — Metabolism is precisely regulated
  - Make only proteins needed at a specific time
- Non-regulated gene expression
  - RNA-poly binds freely to promoter
  - Constitutive genes— Enzymes always needed (e.g., glycolysis)
- **Negative gene regulation**
  - Repressor protein binds operator
    → Block RNA polymerase → Inhibits gene expression
    → Decreases synthesis of enzymes
- **Positive gene regulation**
  - Activator protein binds separate binding site near promoter
    → Enhance RNA polymerase activity
- **Regulon**: multiple operons regulated by the same regulator.
  - >40 regulons identified in *E. coli*

Negative gene regulation

The operon default is usually turned "on"

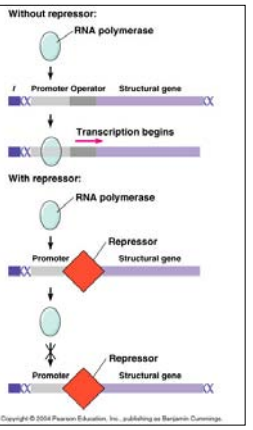
Repressor protein binds operator
→ Block RNA polymerase
→ Inhibits gene expression
→ Decreases synthesis of enzymes



Negative gene regulation

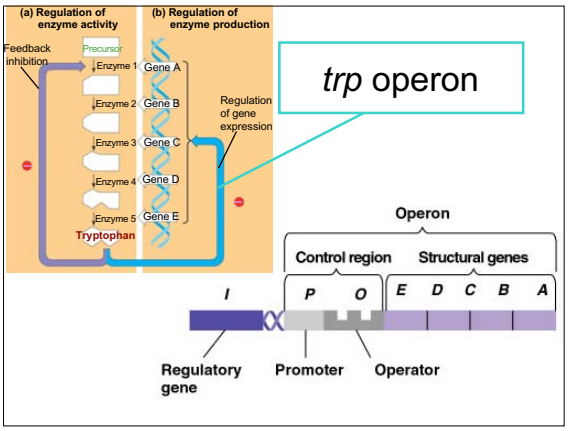
Regulating the regulator:

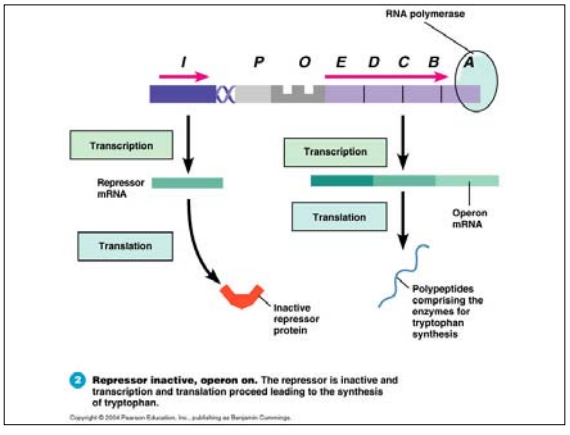
- In a **repressible** operon
  - A **corepressor** molecule binds allosterically to the repressor protein → repressor protein assumes <u>active</u> configuration → binds to the operator → <u>shuts off</u> transcription
- In an **inducible** operon
  - A **inducer** molecule binds allosterically to the repressor protein → repressor protein assumes <u>inactive</u> configuration → cannot bind to the operator → <u>turns on</u> transcription

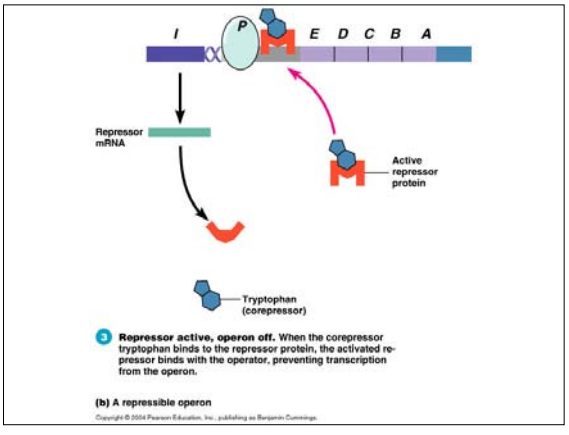


Repressible Operon

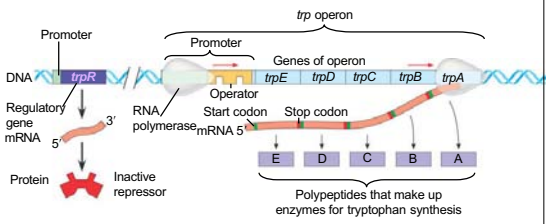
- Tryptophan operon
- Usually occurs in anabolism
- Consists of 5 structural genes
- Repressor is inactive so tryptophan is synthesized
- Amino acid in media
  - Binds to repressor activating it







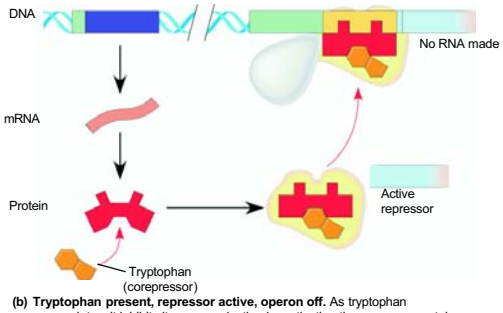
trp operon



(a) Tryptophan absent, repressor inactive, operon on. RNA polymerase attaches to the DNA at the promoter and transcribes the operon's genes.

Figure 18.21a

trp operon



(b) Tryptophan present, repressor active, operon off. As tryptophan accumulates, it inhibits its own production by activating the repressor protein.

Figure 18.21b

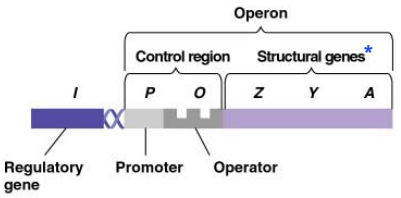
Inducible Operon

- Turn on the transcription of gene
- Inducer- induces transcription
- Inducible enzymes
  - Synthesized only when substrate is present
  - Lactose metabolism in *E. coli*

lac Operon

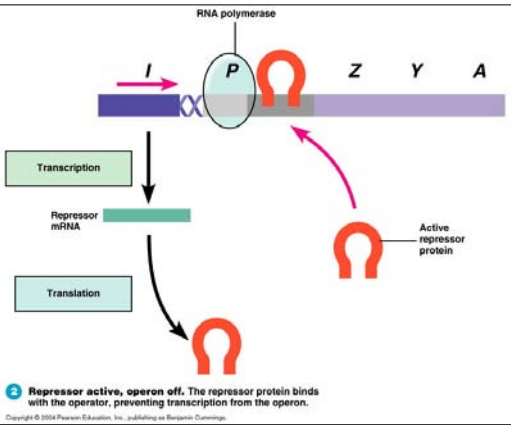
- Inducible operon: enzymes to metabolize lactose
- Regulatory sites
  - Promoter- RNA polymerase
  - Operator- repressor binds
- i genes code for repressor-regulatory protein
  - Outside operon
  - Always turned on (constitutive gene)
  - Binds to operator
- Structural genes
  - *lac* operon-3 genes

lac Operon



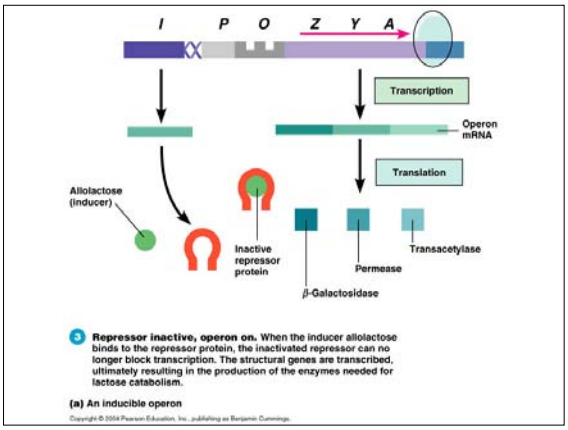
*Genes coding for enzymes to digest and utilize lactose:

Z ⇒ β-galactosidase
Y ⇒ permease
A ⇒ transacetylase

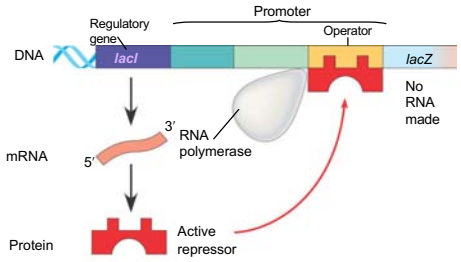


Lactose in Medium

- Binds repressor changing shape
- Repressor can't bind to Operator
- RNA polymerase can bind to Promoter
- Enzymes for lactose metabolism produced
  - Lactose transported into cell
  - Metabolized into glucose and galactose



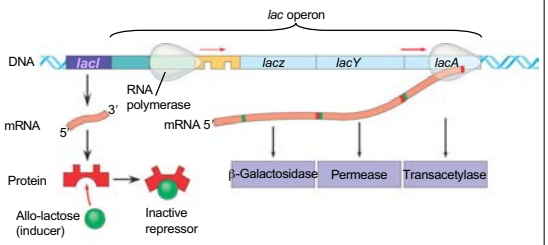
lac Operon



(a) Lactose absent, repressor active, operon off. The *lac* repressor is innately active, and in the absence of lactose it switches off the operon by binding to the operator.

Figure 18.22a

lac Operon



(b) Lactose present, repressor inactive, operon on. Allo-lactose, an isomer of lactose, derepresses the operon by inactivating the repressor. In this way, the enzymes for lactose utilization are induced.

Figure 18.22b

Positive Gene Regulation

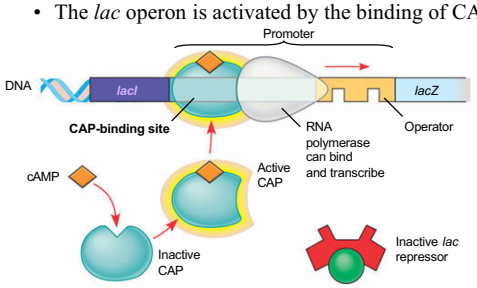
- Some operons are also subject to **positive gene regulation**
  - Stimulatory **activator** protein binds separate binding site near promoter
    → Enhance RNA polymerase activity
    → Increase gene expression & enzyme synthesis
  - **Catabolite Activator Protein** (CAP)
    - Activates many catabolic pathways
    - Including *lac* operon.

Positive regulation of the *lac* operon

- In *E. coli*, when glucose is the preferred energy substrate.
- When available glucose decreases, intracellular cAMP (**c**yclic-**a**denosine **m**ono**p**hosphate) increases.
- cAMP binds to CAP, causing it to change into the active configuration.
- Active CAP enhances operon promoters for alternative catabolic pathways, including the *lac* operon.

Positive regulation of the *lac* operon

- The *lac* operon is activated by the binding of CAP

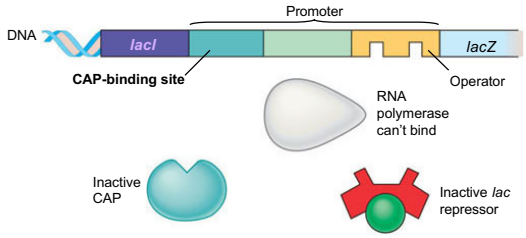


(a) Lactose present, glucose scarce (cAMP level high): abundant *lac* mRNA synthesized.

Figure 18.23a

Positive regulation of the *lac* operon

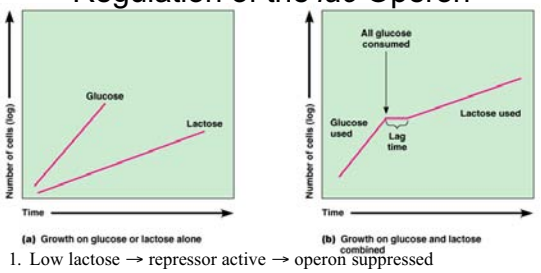
- ↑glucose levels → ↓cAMP → inactive CAP



(b) Lactose present, glucose present (cAMP level low): little *lac* mRNA synthesized.

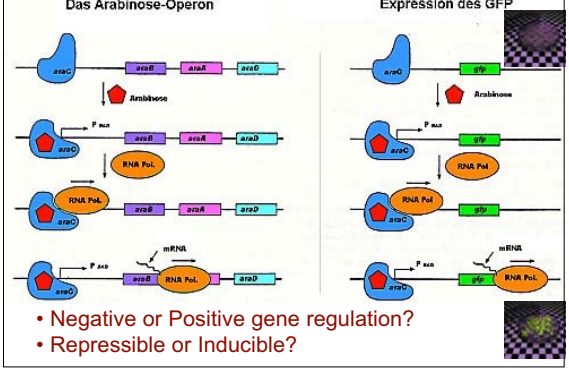
Figure 18.23b

Combined Negative & Positive Regulation of the *lac* Operon



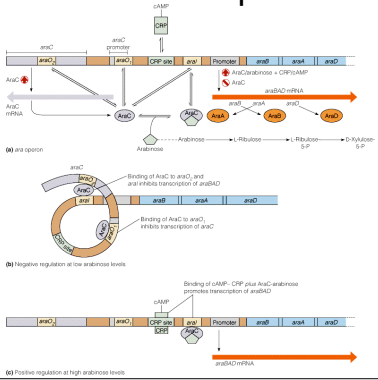
1. Low lactose → repressor active → operon suppressed
2. High lactose/high glucose → repressor inactive but activator inactive → low operon activity
3. High lactose/low glucose → repressor inactive and activator active → high operon activity → enhanced lactose catabolism

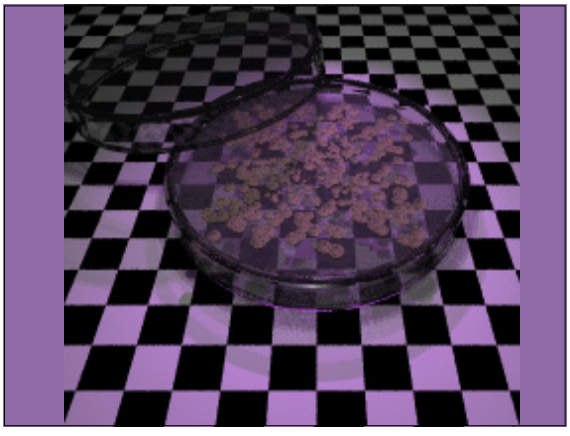
Arabinose Operon



- Negative or Positive gene regulation?
- Repressible or Inducible?

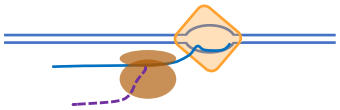
Arabinose Operon



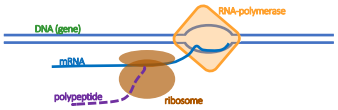


Regulation of operons in prokaryotes by

Transcription Attenuation

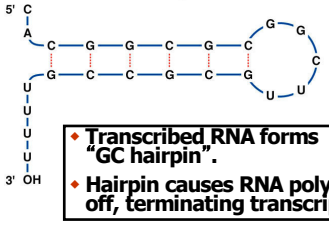


Simultaneous transcription & translation in prokaryotes



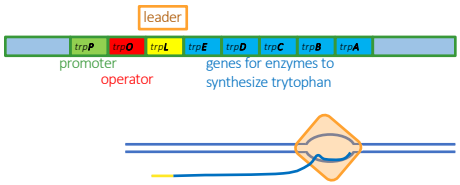
Transcription Termination (prokaryotes)

GC Hairpin



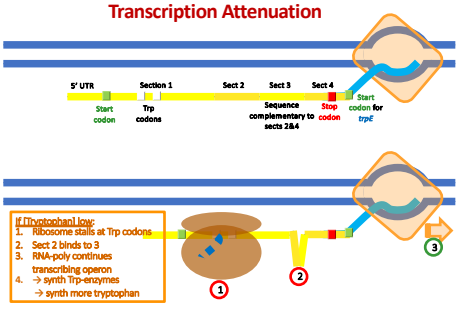
- Transcribed RNA forms "GC hairpin".
- Hairpin causes RNA poly to fall off, terminating transcription.

***trp* operon revisited**



Regulation of operons in prokaryotes by

Transcription Attenuation



Regulation of operons in prokaryotes by

Transcription Attenuation