Why regulate?

- **Waste of energy to make what you don't need**
- protein synthesis is expensive
- have components present in appropriate amounts
- **Adapt to changes in environment**
- change to new nutrient sources-> catabolism
- synthesize new substances -> anabolism
- shutdown production of unnecessary proteins->virulence
- different stages in development= sporulation, biofilm formation
- changes based on "hormones"= quorum sensing
- genes expressed all of the time are **constitutive**

Levels at which to regulate

- RNA
  - Transcription
  - Post-transcriptional
    - mRNA stability
    - Translation of mRNA
- Protein
  - Stability
  - Enzymatic activity

Transcriptional repression

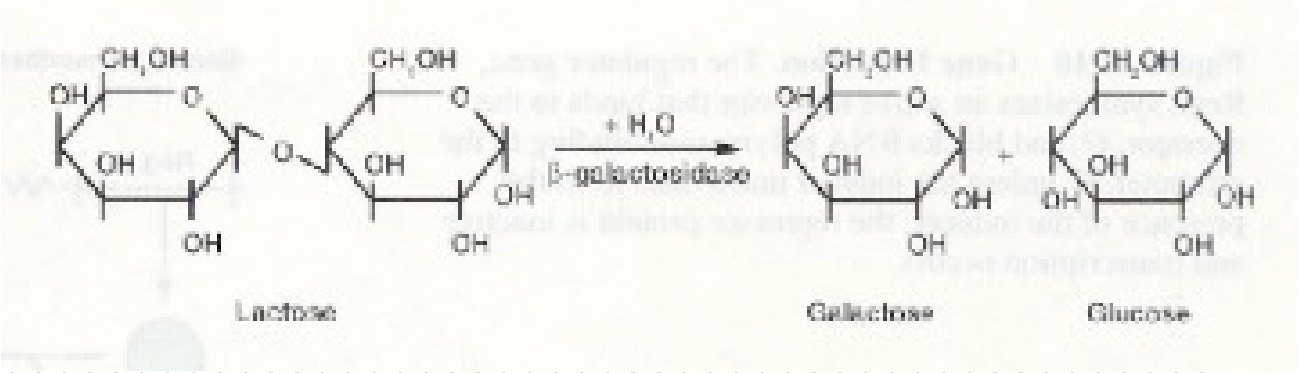
- Repression= negative regulation
  - Inhibition of transcription
  - Usually responsive to large amount of end product of pathway
  - Mediated by DNA binding proteins= repressors
  - Repressors sterically hinder or interact with RNA polymerase to prevent transcription

Transcriptional activation

- Activation of transcription
- Mediated by DNA binding proteins= activators
- Activators may:
  - Interact with RNA polymerase to increase initiation
  - Make DNA more accessible to RNA polymerase

    (e.g. displace repressors, bend DNA)

The *lac* operon



Promoter for regulatory gene

Regulatory gene

Structural loci

Promoter for *lac* operon

P_I *i* P_{lac} *o* *z* *y* *a*

Operator

Structural gene for β-galacto-sidase

Structural gene for β-galactoside permease

Structural gene for β-galactoside transacetylase

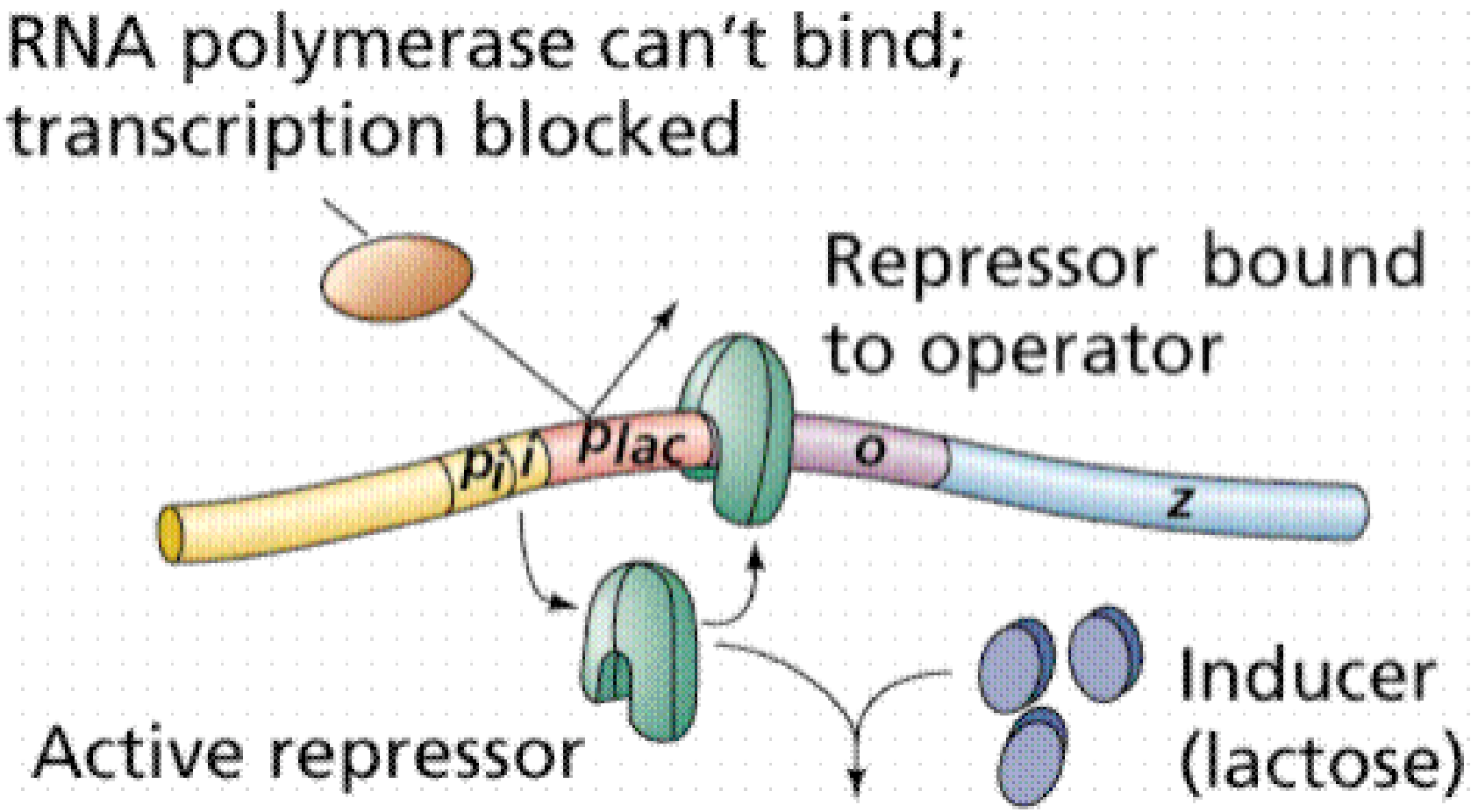
***lac* Operon**

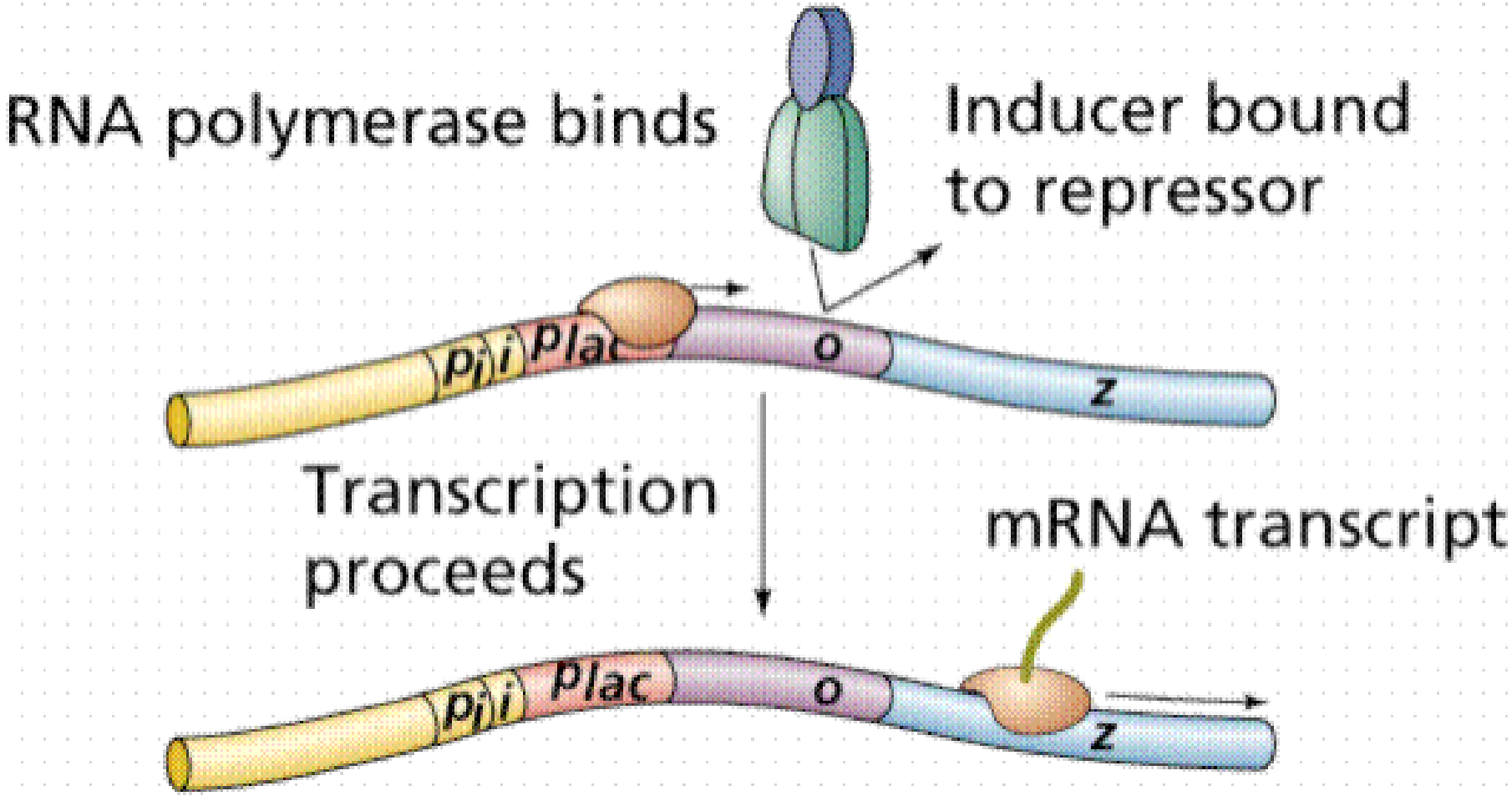
Transcriptional regulatory proteins that control *lac* operon

- Repression
  - LacI repressor binding
- Activation
  - cAMP-CAP activator binding

Repression of the *lac* operon

-occurs in absence of lactose




RNA polymerase binds
Inducer bound to repressor
Transcription proceeds
mRNA transcript
Pi
i
Plac
o
z
Pi
i
Plac
o
z

Diauxic Growth

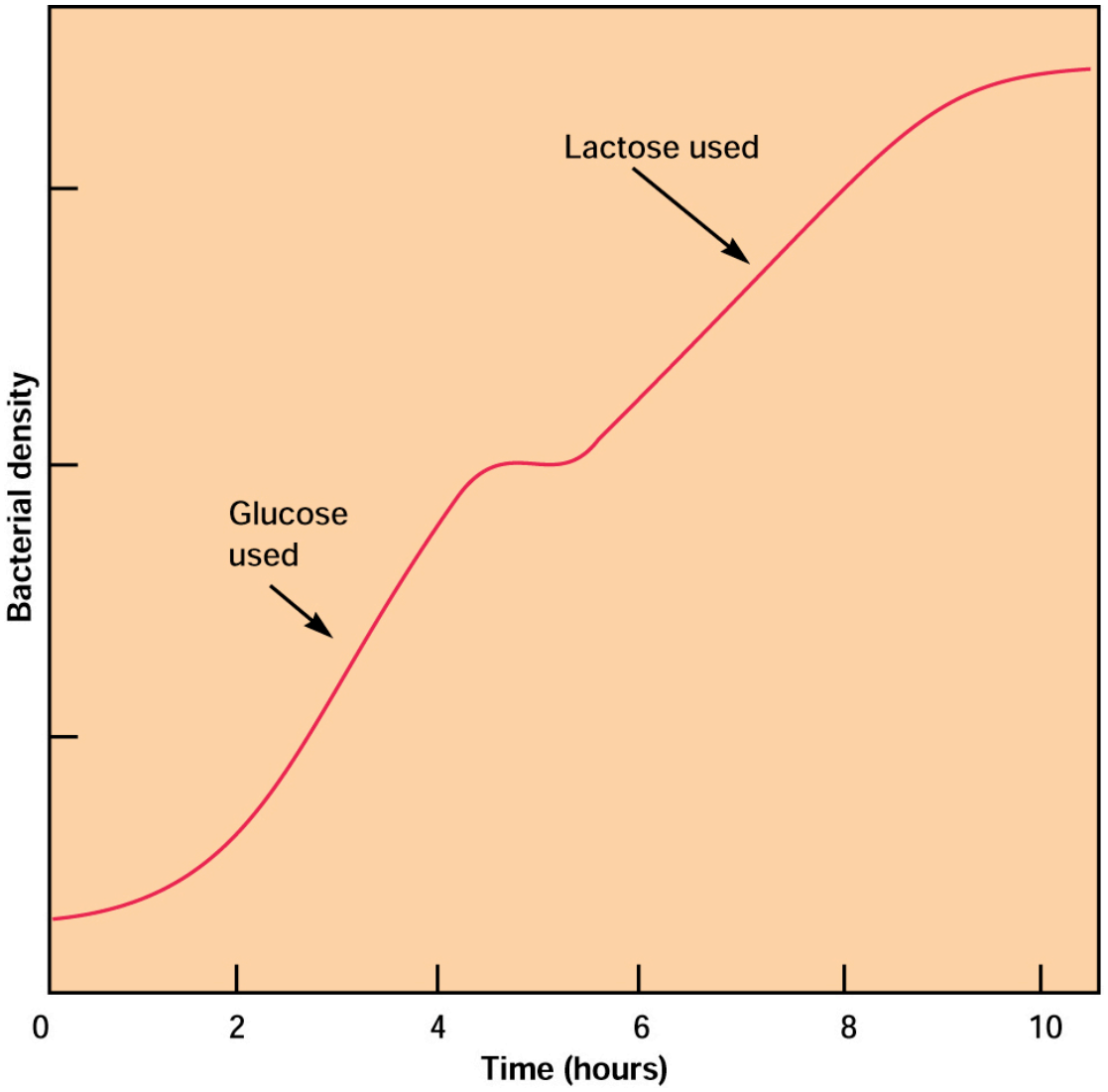


Fig. 12. 31

Response to carbon sources

Glucose present, lactose present

- Bacterium utilizes glucose
- Does not transcribe *lac* operon
  - Repressor (LacI) is inactivated due to allolactose
  - However, no activation due to lack of cAMP-CAP

Glucose absent, lactose present

- Utilize lactose
- Transcribe *lac* operon
  - Repressor (LacI) is inactivated due to allolactose
  - Activation by cAMP-CAP interaction with RNA polymerase

Activation of *lac* operon

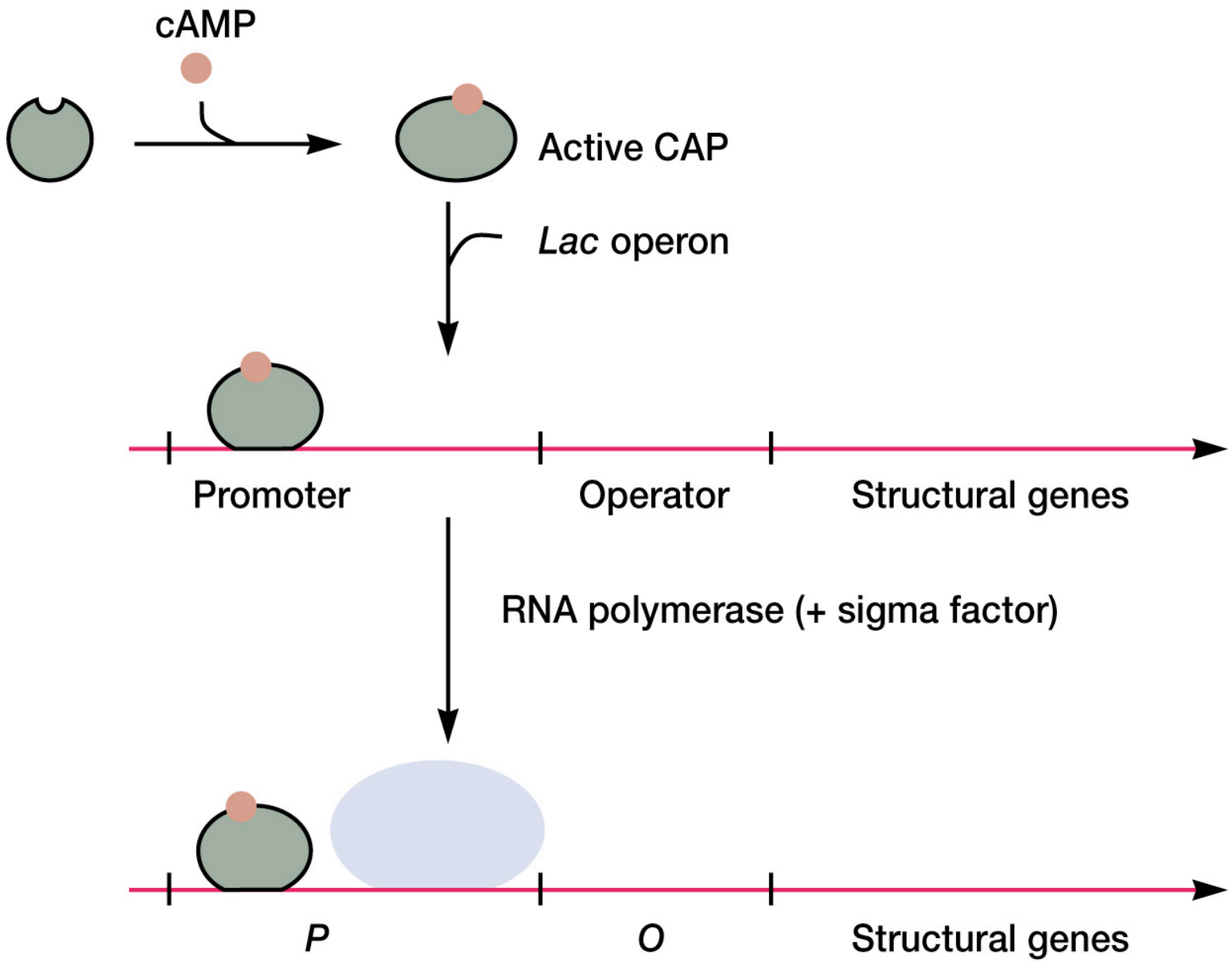


Fig. 12.27

Production of cAMP linked to group translocation PTS system

Enzyme II complex
-donates PO4 to glucose
-activates adenyl cyclase
-lots of glucose, decreased amount of enzyme II-PO4
-decreased active adenyl cyclase

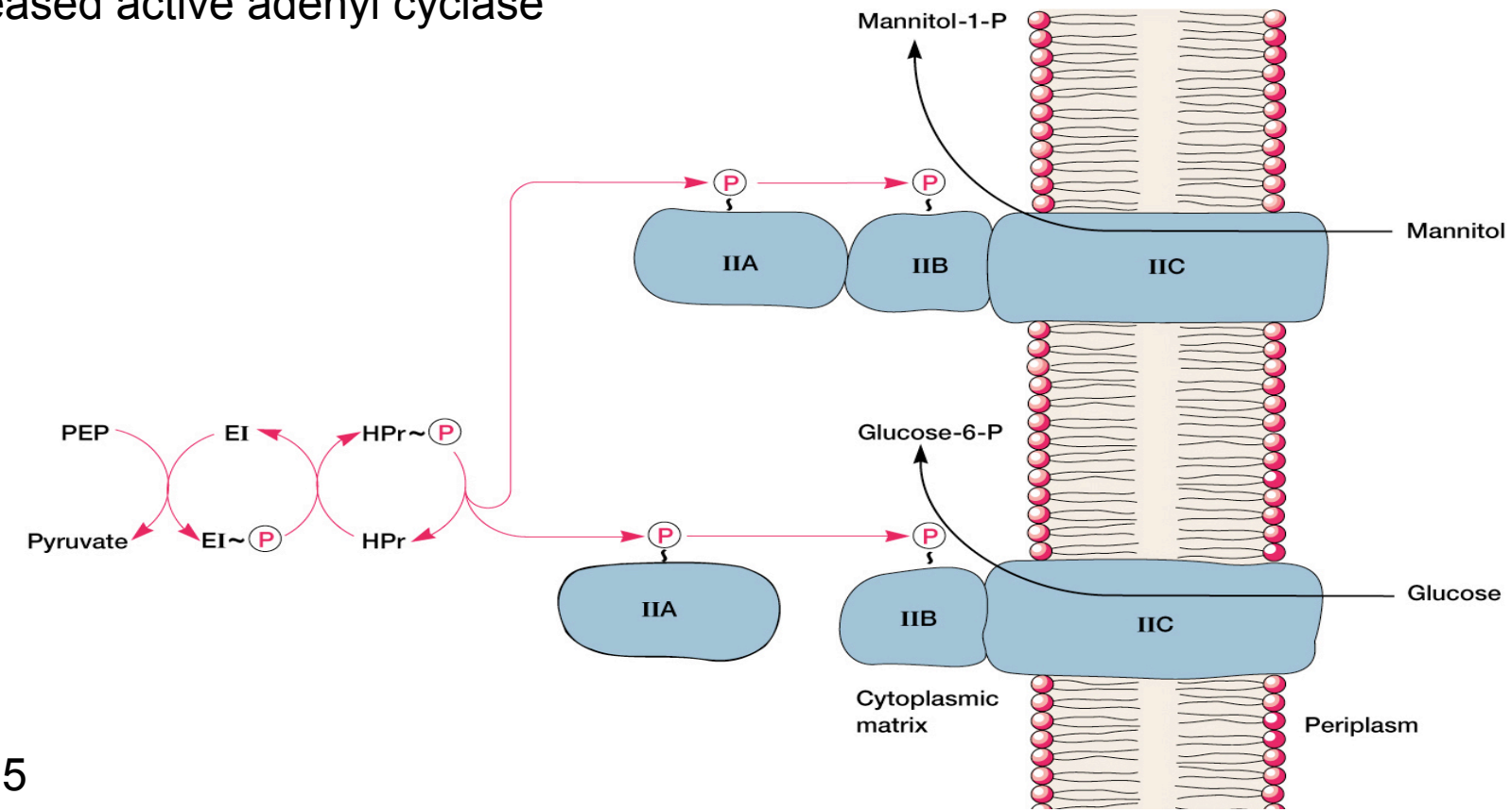
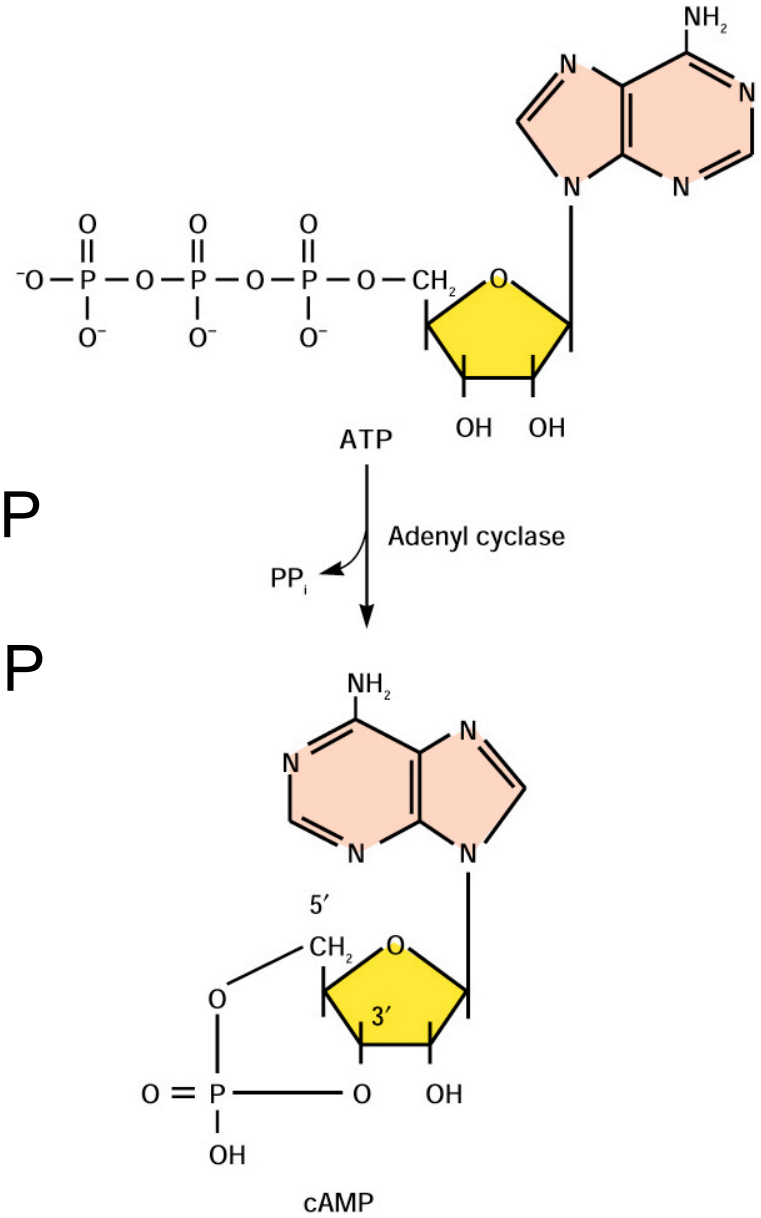


Fig. 5.5

Adenyl cyclase activity linked to PTS system

High glucose= ↓ cAMP

Low glucose= ↑ cAMP



cAMP-CAP

- Complex of cAMP and the protein CAP
- CAP can only bind to DNA when cAMP is bound to it

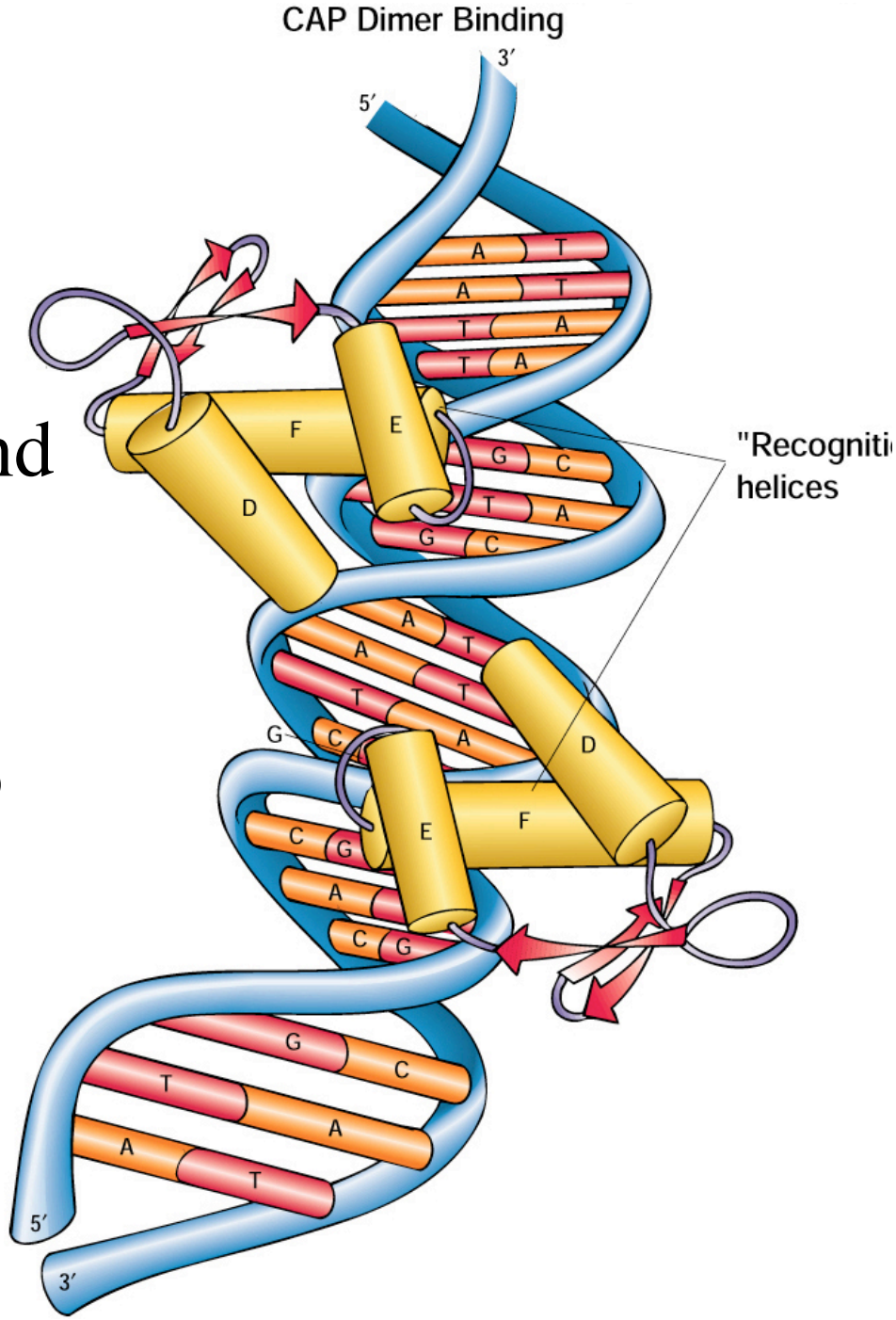


Fig. 12.28

Activation of *lac* operon

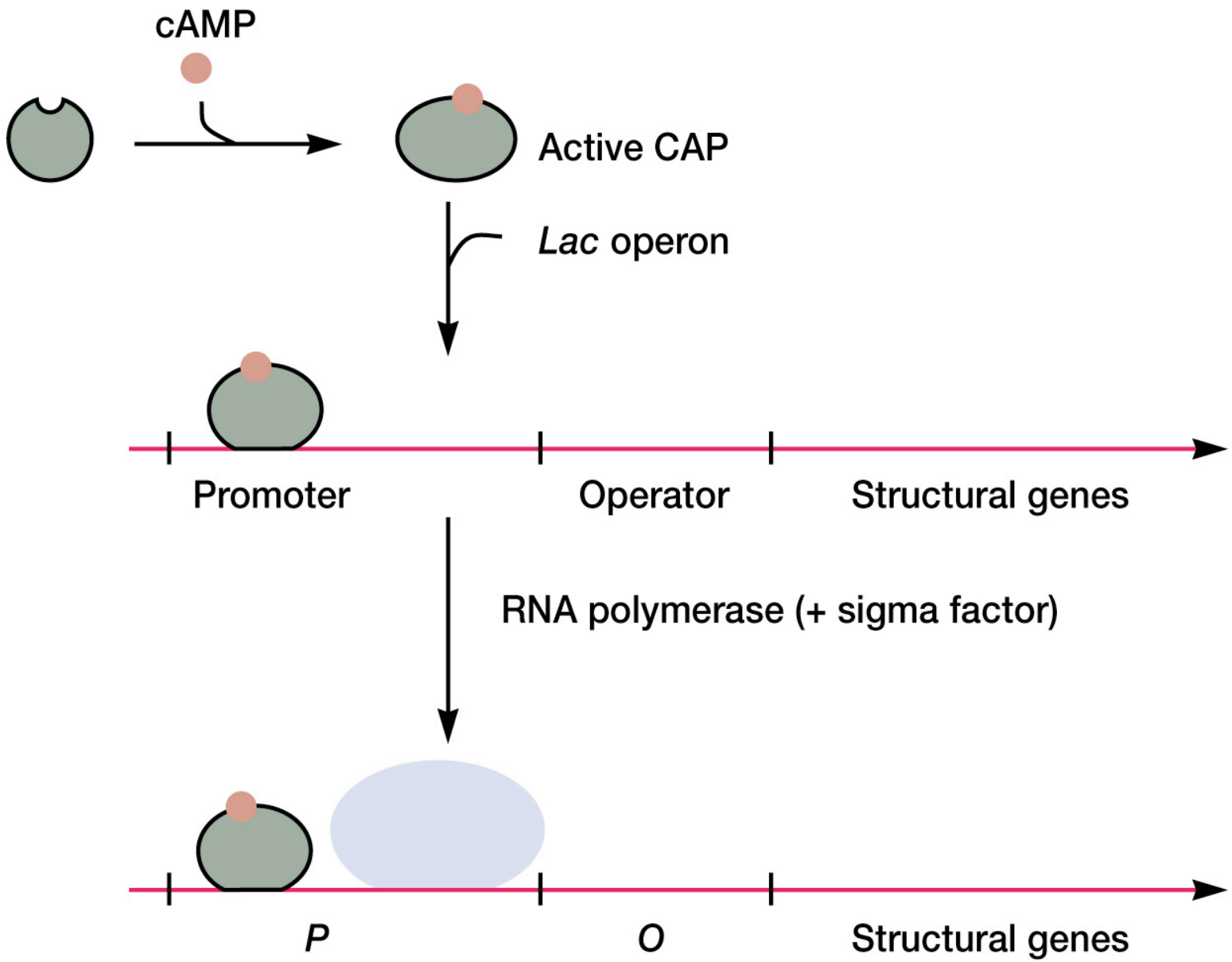
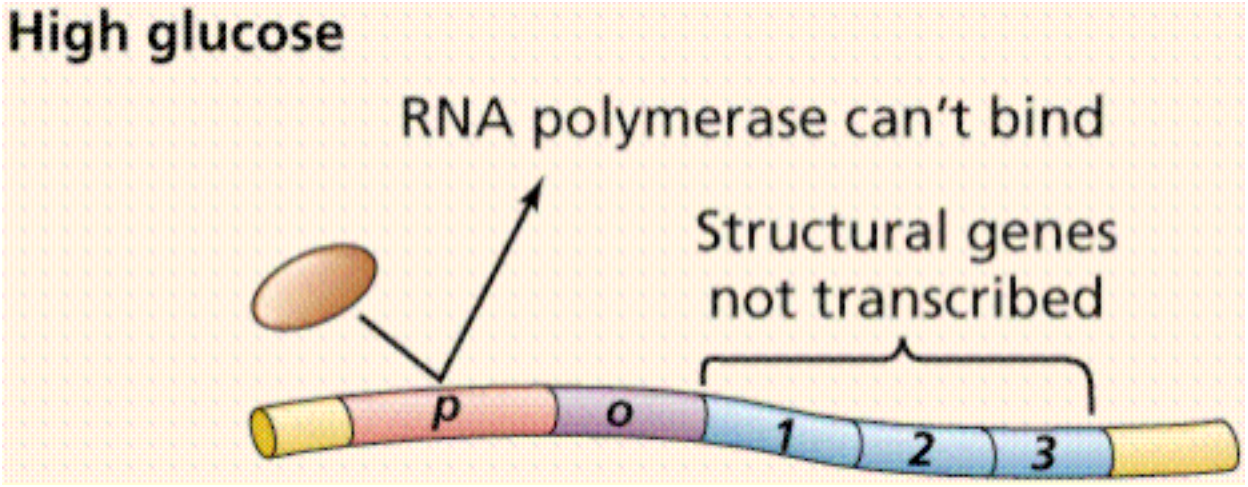
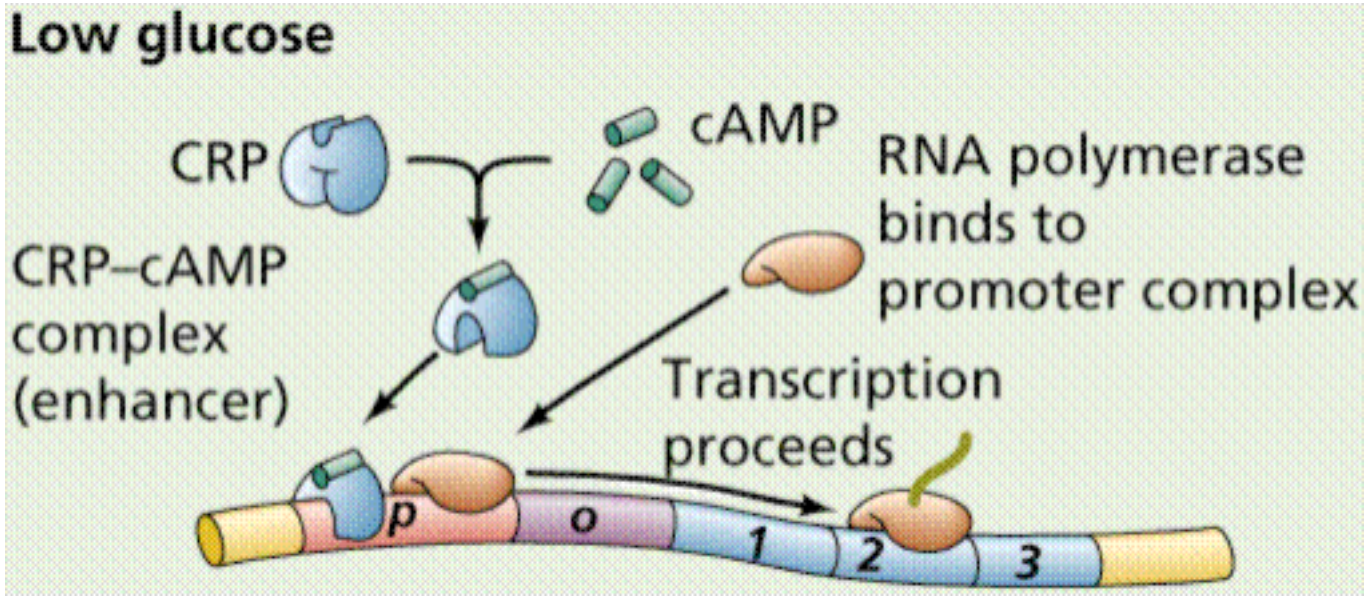


Fig. 12.27

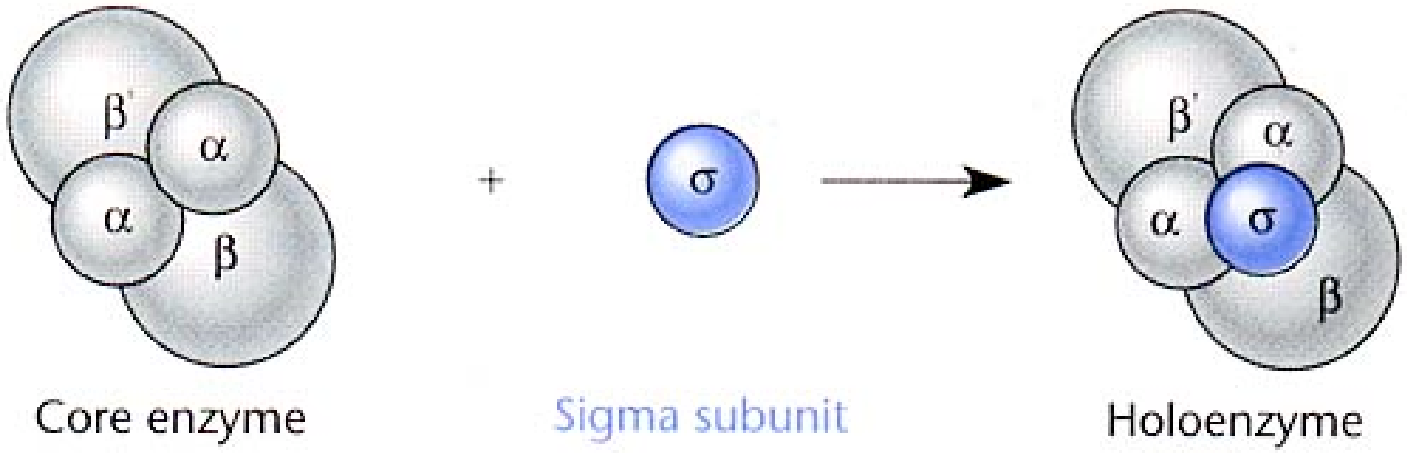
Activation of the *lac* operon





Sigma factors

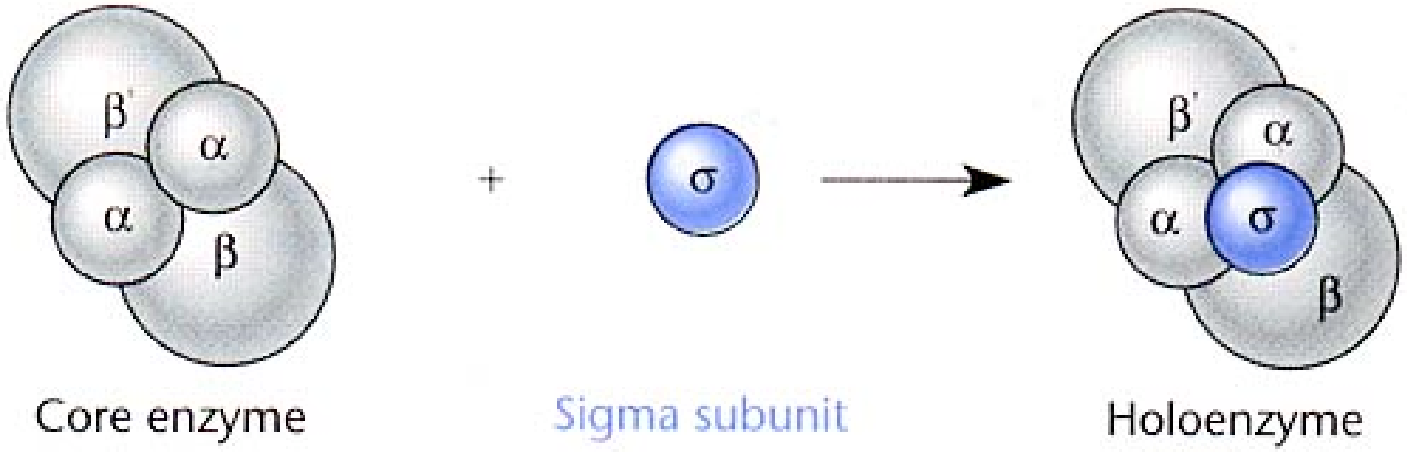
- Recognize promoter in DNA
- Variety of sigma factors in bacteria
- Different sigma factors control different subsets of genes



Sigma factors

- σ^{70} =housekeeping genes
- σ^{32} = heat shock
- σ^{28} = chemotaxis/flagella
- $\sigma^{F/}$ σ^{G} = sporulation
- σ^{E} = extracytoplasmic stress
- σ^{S} = general stress response

σ^S mediates expression of general stress response



Multiple cues control σ^S expression leading to global gene response

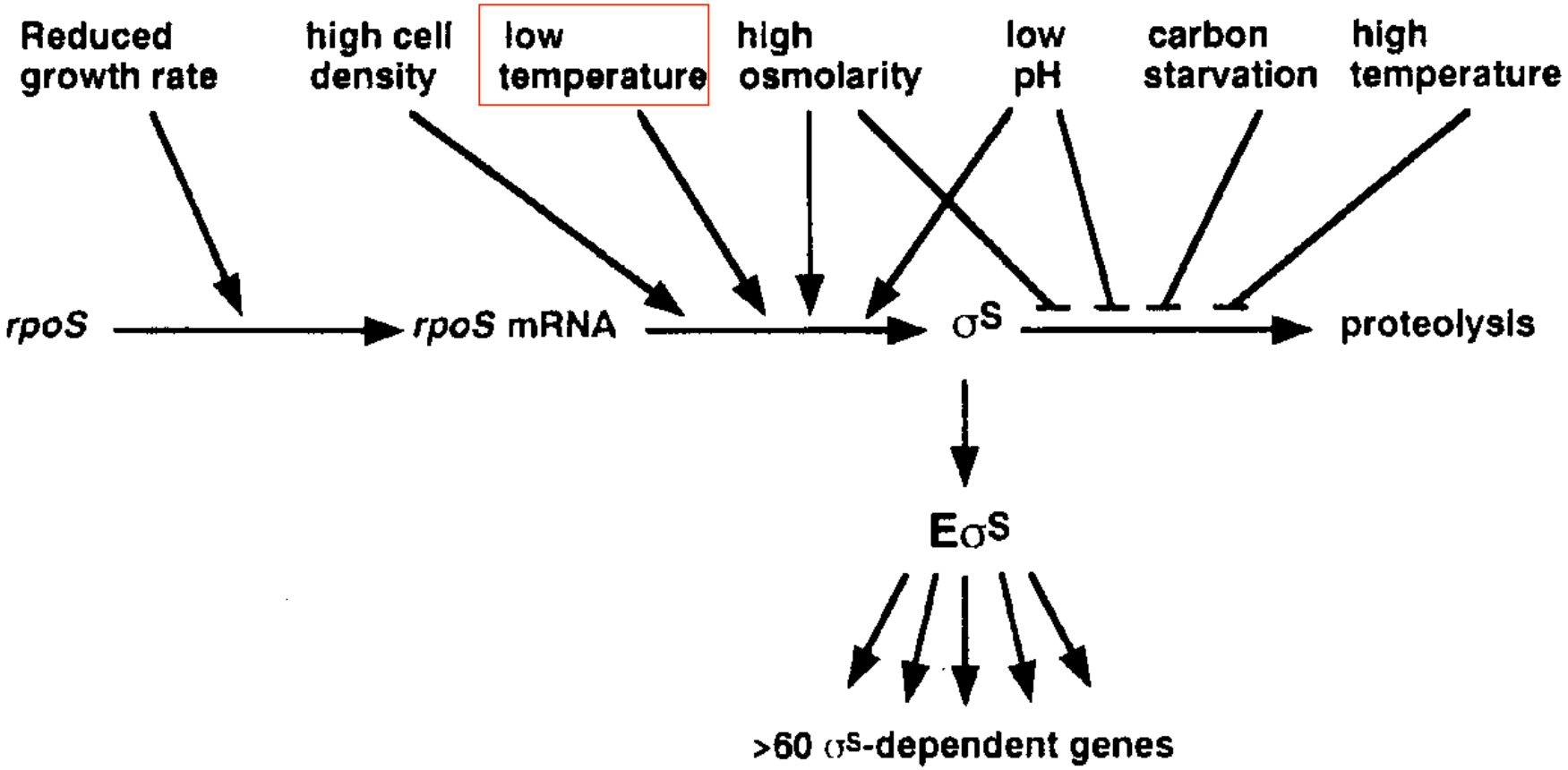
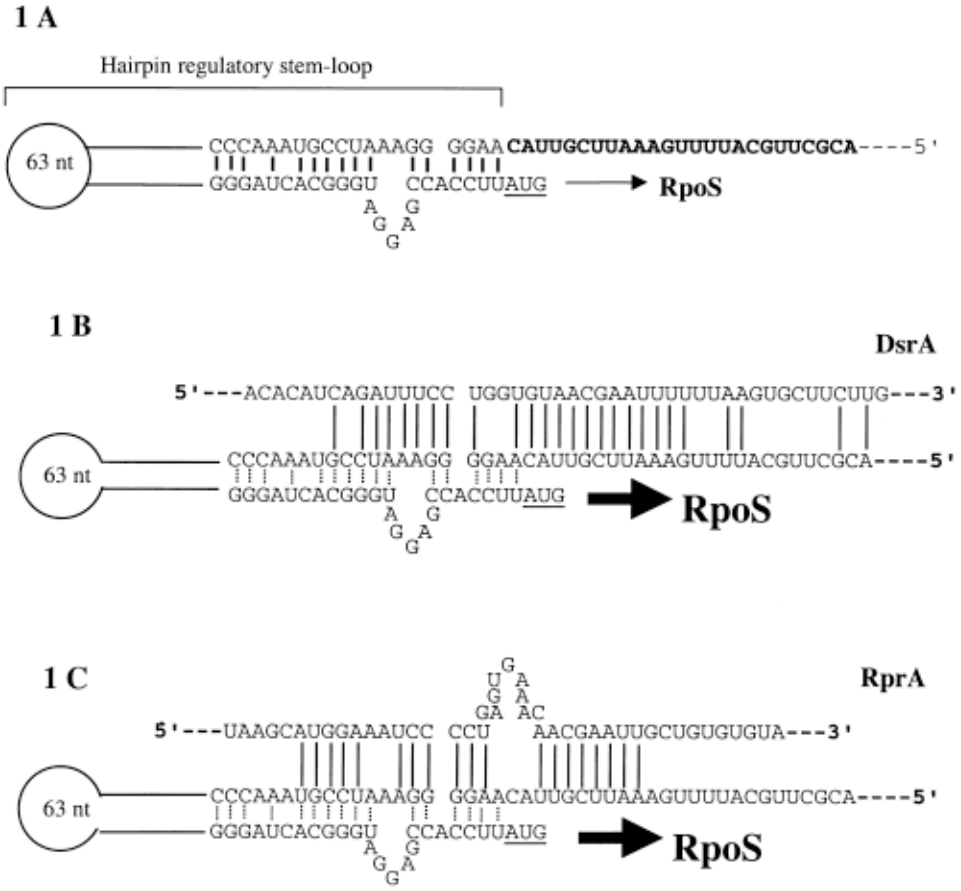


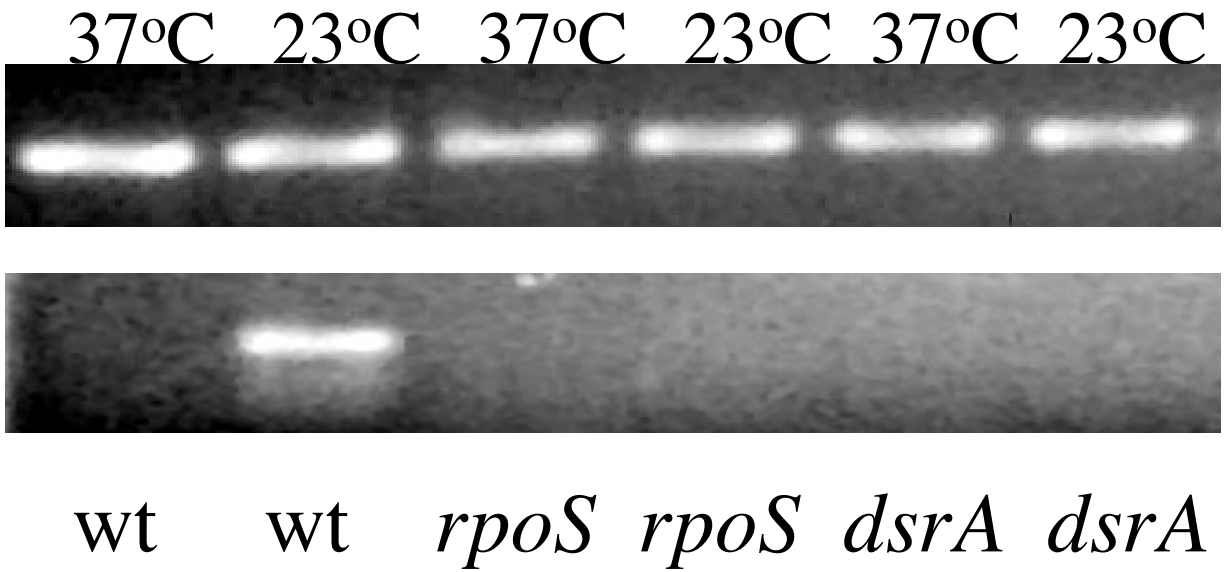
Figure from *Bacterial Stress Responses* 2000. Storz and Hengge-Aronis, ed.

Small sRNAs

- Gene product is the RNA
- sRNAs are not translated
- 50-400 bp in length
- Serve a variety of regulatory functions
  - Oxidative stress (OxyS)
  - Iron acquisition (RhyB)
  - Porin synthesis (MicF)
  - Sigma S (DsrA)
- Act in a variety of ways
  - Alter mRNA stability (RhyB)
  - Alter translation (OxyS, DsrA, MicF)

dsrA increases σ^S translation





- *rimJ*
- *csgA*

•*rimJ*=housekeeping gene
•*rpoS* required for *csgA* transcription
•*dsrA* required for increased transcription of *csgA* at low temperature

Levels at which to regulate

- RNA
  - Transcription (*lac* operon, sigma factors)
  - Post-transcriptional
    - mRNA stability (RhyB)
    - Translation of mRNA (sigma S)

- Protein
  - Stability (sigma S)
  - Enzymatic activity (chemotaxis, cAMP production)