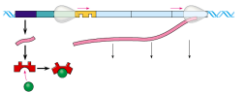
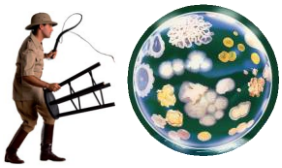
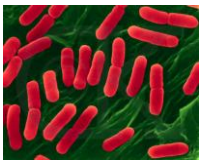


Control of Prokaryotic (Bacterial) Genes



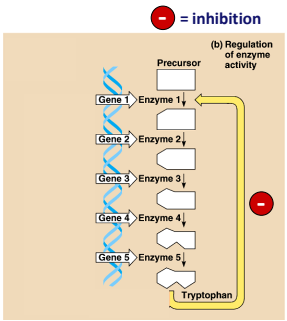


Bacterial metabolism

- Bacteria need to respond quickly to changes in their environment
  - if they have enough of a product, need to stop production
    - why? waste of energy to produce more
    - how? stop production of enzymes for synthesis
  - if they find new food/energy source, need to utilize it quickly
    - why? metabolism, growth, reproduction
    - how? start production of enzymes for digestion

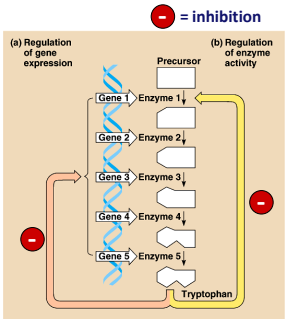
Remember Regulating Metabolism?

- Feedback inhibition
  - product acts as an allosteric inhibitor of 1st enzyme in tryptophan pathway
  - *but this is wasteful production of enzymes*



Different way to Regulate Metabolism

- Gene regulation
  - instead of blocking enzyme function, block transcription of genes for all enzymes in tryptophan pathway
    - saves energy by not wasting it on unnecessary protein synthesis



Gene regulation in bacteria

- Cells can vary the amount of specific enzymes by regulating gene transcription
  - turn genes on or turn genes off
    - turn genes OFF example

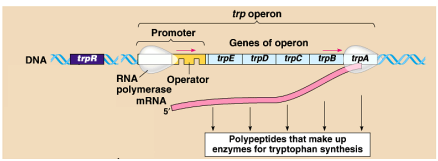
if bacterium has enough tryptophan then it doesn't need to make enzymes used to build tryptophan

- turn genes ON example

if bacterium encounters new sugar (energy source), like lactose, then it needs to start making enzymes used to digest lactose

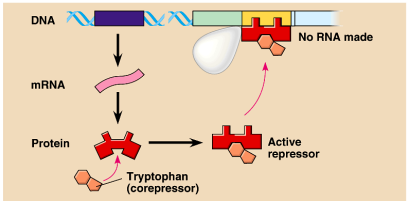
Bacteria group genes together

- Operon
  - genes grouped together with related functions
    - example: all enzymes in a metabolic pathway
  - promoter = RNA polymerase binding site
    - single promoter controls transcription of all genes in operon
    - transcribed as one unit & a single mRNA is made
  - operator = DNA binding site of repressor protein



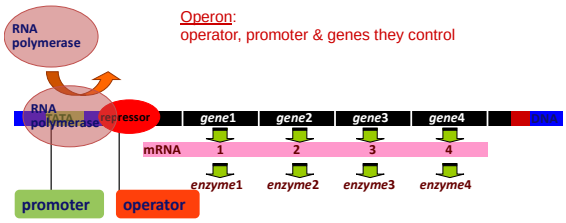
So how can these genes be turned off?

- Repressor protein
  - binds to DNA at operator site
  - blocking RNA polymerase
  - blocks transcription



Operon Model

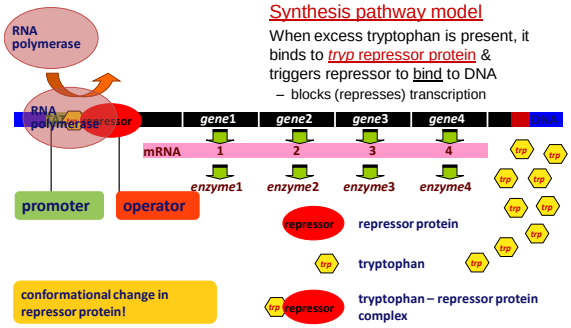
Operon:
operator, promoter & genes they control



Repressor protein turns off gene by blocking RNA polymerase binding site.

repressor = repressor protein

Repressible operon: tryptophan



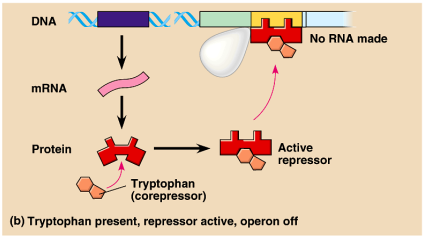
Synthesis pathway model

When excess tryptophan is present, it binds to *trp* repressor protein & triggers repressor to bind to DNA
- blocks (represses) transcription

Tryptophan operon

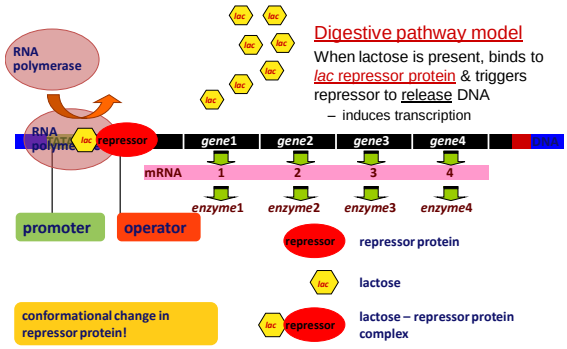
What happens when tryptophan is present?

Don't need to make tryptophan-building enzymes



Tryptophan is allosteric regulator of repressor protein

Inducible operon: lactose



Digestive pathway model

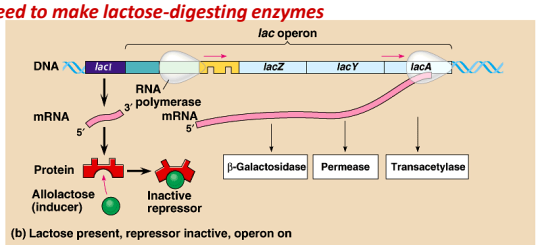
When lactose is present, binds to *lac* repressor protein & triggers repressor to release DNA

- induces transcription

Lactose operon

What happens when lactose is present?

Need to make lactose-digesting enzymes

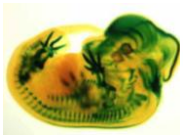


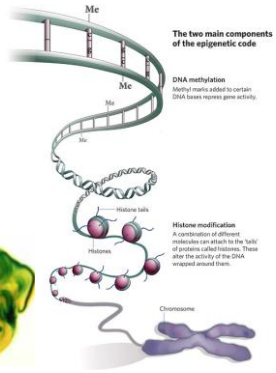
Lactose is allosteric regulator of repressor protein

Operon summary

- Repressible operon (Trp Operon)
  - usually functions in anabolic pathways
    - synthesizing end products
  - when end product is present in excess, cell allocates resources to other uses
- Inducible operon (Lac Operon)
  - usually functions in catabolic pathways,
    - digesting nutrients to simpler molecules
  - produce enzymes only when nutrient is available
    - cell avoids making proteins that have nothing to do, cell allocates resources to other uses

Control of Eukaryotic Genes





The BIG Questions…

- How are genes turned on & off in eukaryotes?
- How do cells with the same genes differentiate to perform completely different, specialized functions?

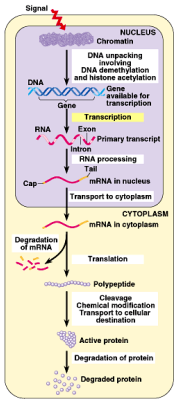
(a) 5 weeks.

(b) 14 weeks.

(c) 20 weeks.

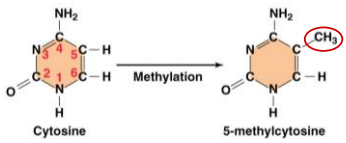
Points of control

- The control of gene expression can occur at any step in the pathway from gene to functional protein
  1. packing/unpacking DNA
  2. transcription
  3. mRNA processing
  4. mRNA transport
  5. translation
  6. protein processing
  7. protein degradation



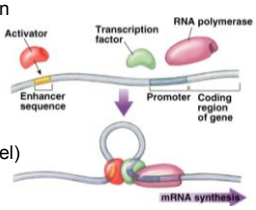
DNA methylation

- Methylation of DNA blocks transcription factors
  - no transcription
    → genes turned off
  - attachment of methyl groups ($-CH_3$) to cytosine
    - C = cytosine
  - nearly permanent inactivation of genes
    - ex. inactivated mammalian X chromosome = Barr body

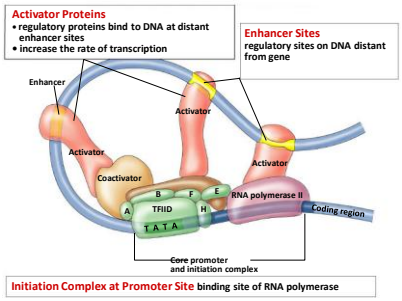


Transcription initiation

- Control regions on DNA
  - promoter
    - nearby control sequence on DNA
    - binding of RNA polymerase & transcription factors
    - "base" rate of transcription
  - enhancer
    - distant control sequences on DNA
    - binding of activator proteins
    - "enhanced" rate (high level) of transcription

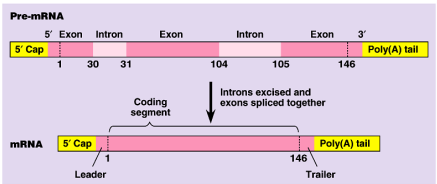


Transcription complex



Regulation of mRNA degradation

- 5' cap and Poly-A Tail
- Life span of mRNA determines amount of protein synthesis
  - mRNA can last from hours to weeks



RNA interference

- Small interfering RNAs (siRNA)
  - short segments of RNA (21-28 bases)
    - bind to mRNA
    - create sections of double-stranded mRNA
    - "death" tag for mRNA
      - triggers degradation of mRNA
  - cause gene "silencing"
    - post-transcriptional control
    - turns off gene = no protein produced

siRNA

1990s | 2006

RNA interference

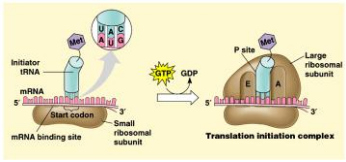
Andrew Fire — Stanford
Craig Mello — U Mass

Nobel Prize

"for their discovery of RNA interference — gene silencing by double-stranded RNA"

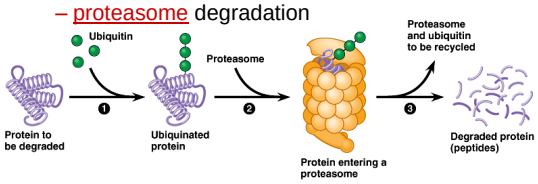
Control of translation

- Block initiation of translation stage
  - regulatory proteins attach to 5' end of mRNA
    - prevent attachment of ribosomal subunits & initiator tRNA
    - block translation of mRNA to protein



Protein processing & degradation

- Protein processing
  - folding, cleaving, adding sugar groups, targeting for transport
- Protein degradation
  - ubiquitin tagging
  - proteasome degradation



1980s | 2004

Ubiquitin

- "Death tag"
  - mark unwanted proteins with a label
  - 76 amino acid polypeptide, ubiquitin
  - labeled proteins are broken down rapidly in "waste disposers"
    - proteasomes

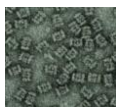

Aaron Ciechanover
Israel


Avram Hershko
Israel

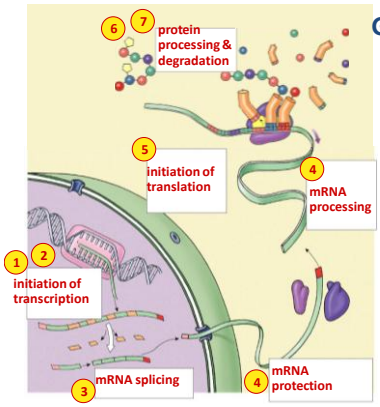

Irwin Rose
UC Riverside

Proteasome

- Protein-degrading "machine"
  - cell's waste disposer
  - breaks down any proteins into 7-9 amino acid fragments
    - cellular recycling



play Nobel animation



Gene Regulation

1 & 2. transcription
- DNA packing
- transcription factors

3 & 4. post-transcription
- mRNA processing
- splicing
- 5' cap & poly-A tail
- breakdown by siRNA

5. translation
- block start of translation

6 & 7. post-translation
- protein processing
- protein degradation