CHAPTER 31: CONTROL OF GENE EXPRESSION

SELECTED LECTURE TOPICS

**Differential gene expression:** prokaryotes and eukaryotes

**Regulation of gene expression:**

  *Transcription* regulated by DNA-binding proteins

  *E. coli* lactose (lac) operon
  Helix-turn-helix motif: prokaryotic DNA-binding proteins

**Eukaryotic gene regulation:**

  Transcriptional activation/repression is mediated by protein–DNA and/or protein-protein interactions
### Evidence of Eukaryotic Differential Gene Expression:
Relative amounts (in %) of mRNAs

#### TABLE 31.1

<table>
<thead>
<tr>
<th>Rank</th>
<th>Pancreas</th>
<th>%</th>
<th>Liver</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Procarboxypeptidase A1</td>
<td>7.6</td>
<td>Albumin</td>
<td>3.5</td>
</tr>
<tr>
<td>2</td>
<td>Pancreatic trypsinogen 2</td>
<td>5.5</td>
<td>Apolipoprotein A-I</td>
<td>2.8</td>
</tr>
<tr>
<td>3</td>
<td>Chymotrypsinogen</td>
<td>4.4</td>
<td>Apolipoprotein C-I</td>
<td>2.5</td>
</tr>
<tr>
<td>4</td>
<td>Pancreatic trypsin 1</td>
<td>3.7</td>
<td>Apolipoprotein C-III</td>
<td>2.1</td>
</tr>
<tr>
<td>5</td>
<td>Elastase IIIB</td>
<td>2.4</td>
<td>ATPase 6/8</td>
<td>1.5</td>
</tr>
<tr>
<td>6</td>
<td>Protease E</td>
<td>1.9</td>
<td>Cytochrome oxidase 3</td>
<td>1.1</td>
</tr>
<tr>
<td>7</td>
<td>Pancreatic lipase</td>
<td>1.9</td>
<td>Cytochrome oxidase 2</td>
<td>1.1</td>
</tr>
<tr>
<td>8</td>
<td>Procarboxypeptidase B</td>
<td>1.7</td>
<td>α-1-Antitrypsin</td>
<td>1.0</td>
</tr>
<tr>
<td>9</td>
<td>Pancreatic amylase</td>
<td>1.7</td>
<td>Cytochrome oxidase 1</td>
<td>0.9</td>
</tr>
<tr>
<td>10</td>
<td>Bile salt-stimulated lipase</td>
<td>1.4</td>
<td>Apolipoprotein E</td>
<td>0.9</td>
</tr>
</tbody>
</table>

1,000 ................................. 0.001 (or less)
Theme: Control involves sequence-specific DNA binding proteins and/or protein-protein interactions.

Phage repressor protein bound to target sequence: “operator”
Control of Prokaryotic (Differential) Gene Expression:

Example: Transcriptional regulation of the lactose operon
E. coli cells can use lactose as carbon source for growth
β-galactosidase is an inducible enzyme

Lactose removed

(-) inducer

[~ 7% of cell’s total protein]

(+ ) inducer

β-Galactosidase (µg)

Lactose added

Total bacterial protein (µg)
(A) Features of an “operon” map: Inducible gene expression
(B) Lactose operon map: A specific inducible operon

(A) Regulator gene

Control sites

Structural genes

(B) Lactose operon

Promoters

Operator

β-galactosidase gene
Lactose operon regulation:

**lac repressor** a (-) regulator

**Inducer:** (+) regulator
Identifying the lactose operator: “footprint” and sequence DNA

Operator DNA

Repressor

DNA fragment

Operator site

Pancratic deoxyribonuclease

Protected operator site

(23 base pairs protected)

lac repressor binds here

[Colors show 2-fold symmetry]

lac operator is a unique sequence

[one lac vs many pur operators]
Lac repressor is a (-) regulator.
*lac operon promoter* differs from typical *E. coli* promoter.

[Diagram showing the comparison between *lac* and typical *E. coli* promoters.]

- **[lac promoter]**
  - Low RNA polymerase affinity
  - Bad

- **[Typical E. coli promoter]**
  - Good
  - high RNA polymerase affinity
CAP protein (cAMP binding protein): a (+) regulator

CAP-DNA complex:
DNA bends 94°

Repressor and CAP protein overlap with RNA polymerase

*** Repressor and CAP protein overlap with RNA polymerase
Repressor and corepressors: Regulation of the Tryptophan (trp) Operon
Helix-turn-helix motif of DNA binding regulatory proteins

Exception: $\beta$-strand interaction is basis of Met operon repressor DNA binding.

34 Angstroms = 1 full turn of DNA helix
Eukaryotic Chromosomes
Linear, organized and complex – not just a DNA molecule
Levels of Complexity of Chromosome Structure

- **Chromosome Pair**
  - Two chromatids (2 × 10 coils)
  - One coil (30 rosettes)
  - One rosette (6 loops)
  - One loop (50 × 10^6 bp)
  - 30 nm Fiber
  - "Beads-on-a-string" form of chromatin

- **Nuclear scaffold proteins**

- **Metaphase**
  - (1000:1 packing ratio)

- **Solenoid**
  - (40:1 packing ratio)

- **Nucleosome**
  - (6-7:1 packing ratio)

- **DNA**
Puffs are actively transcribed regions of chromosomes

DNase sensitive

Under-methylated cytosines

Regulation of Eukaryotic Gene Transcription
Eukaryotic Transcription is Regulated and Mediated by:

- DNA-Protein Interactions
- Protein-protein interactions (see Ch. 29 also)

**EXAMPLES**

**Enhancers:**
- Muscle creatine kinase gene enhancers

**Enhancers and Activators:**
- Steroid hormone responsive genes
Enhancer/activator protein complexes interacts from a distance with RNA polymerase (at promoters):

(Result is efficient initiation and enhanced transcription)
Enhancers: Muscle creatine kinase gene

3 different enhancer sequences
- 3 different activator proteins [one for each enhancer sequence]

Transcribed from creatine kinase promoter

Sets of muscle cells expressing β-galactosidase [blue from X-gal]
Steroid Hormone - Enhancer System (see Ch.29)

1. Receptor binding to RNA polymerase and transcription initiation.

2. Hormone-receptor complex activates the enhancer.

3. Activated enhancer binds to the promoter, leading to mRNA synthesis.

Inactive gene

Enhancer

Promoter

+1 Start site

Active gene

Activated enhancer

Promoter

mRNA synthesis

Binding to RNA Polymerase and transcription initiation
HRE’s: Hormone Receptor elements:
Receptor binding sites on DNA (enhancers)
Steroid-like hormone receptors: Conserved domains

- Binds transcription factors
- Activation (variable)
- DNA binding
- Hormone binding
- ER (Estrogen)
- PR (Progesterone)
- GR (Glucocorticoid)
- VDR (Vitamin D3)
- RAR (Retinoic Acid)
- TR (Thyroxine)
Different combinations of activators switch on different sets of genes.

**EUKARYOTIC TRANSCRIPTION ACTIVATORS**

**Ligand binding**
[Ex: Steroid hormones]

DNA Binding domain

Activation domain

Interacts with other proteins to regulate transcription

DNA
Hormone Receptor - DNA interactions

Activation domain

NH₂

DNA-binding domain

Ligand-binding domain

COOH
Estrogen receptor is activated by hormone binding followed by coactivator binding, etc.
Receptor activation domain and Mediator protein complex interacts with RNA polymerase (at promoters) to turn on transcription.