I. Chapter 19 Control of Gene Expression in Eukaryotes

- **Differential gene expression**: responsible for creating different cell types, arranging them into tissues, and coordinating their activity to form a multicellular society that we call an individual.

A. 19.1 Gene Regulation in Eukaryotes: an Overview

- **Chromatin**: DNA and protein.
- In order for RNA polymerase to bind to the promoter, *chromatin remodeling* must occur before transcription.
- Level of regulation unique to Eukaryotes is RNA processing, or the steps required to produce a mature mRNA from a primary RNA transcript.
- Eukaryotic gene expression can be controlled at:
    - 1. Chromatin remodeling
    - 2. Transcription
    - 3. RNA processing
    - 4. mRNA stabilization
    - 5. Translation
    - 6. Post-translational modification
    - 1-3 occur in the nucleus, 4-6 occur in the cytoplasm.

B. 19.2 Chromatin Remodeling

- **Histone**: DNA associated proteins.
- **Nucleosomes**: the DNA, wrapped around the histones.
- **H1 histone**: attaches to the "linker DNA" and histones. Seals DNA to nucleosome.
- *30nm fiber* attaches to *scaffold proteins* and the entire assembly is folded in a highly condensed way.
- Chromatin must uncondense to expose a promoter so that RNA polymerase can bind to it.
- **DNases** are enzymes that cut DNA
- In normal default states, genes are turned off.
- When DNA is wrapped in the 30nm fiber, the "parking break" is on.
- **DNA methyltransferases**: add methyl groups to cytosine residues.
- C next to G: **CpG**
- Methylated CpG sequences are recognized by proteins that trigger chromatin condensation.
- Activated genes usually have low levels of methylated CpG near their promoter.
- Non-transcribed genes usually have high levels of methylated CpGs near their promoter.
- **Histone code**: particular combinations of histone modifications set the state of chromatin condensation for a particular gene.
- **Histone acetyltransferases (HATs):** add acetyl groups to positively charged lysine residues in histones.
- **Histone deacetylases (HDACs):** removed acetyl groups on positively charged lysine residues in histones.
- Acetylation of histone usually results in uncondensed chromatin- a state associated with active transcription.

- **Chromatin remodeling complexes**: macromolecular machines that harness the energy in ATP to reshape chromatin.
- Chromatin remodeling complexes cause the nucleotides to slide along DNA or knock the histones off.
- DNA methylation, histone modification, and chromatin remodeling complexes work together to fine tune chromatin condensation at specific genes.
- **Epigenetic inheritance**: patterns of inheritance that are due to something other than difference in DNA sequence.
- Muscle cells are different from brain cells not because they have different genes, but largely because they contain different patterns of DNA methylation and histone modifications during the course of their development.

C. 19.3 Initiating Transcription, Regulatory Sequences, and Proteins

- **Tatabox**; binding site on DNA.
- **Tata binding proteins (TBP):** proteins that bind to the tata box.
- **Regulatory sequences**: any sequence of DNA that is involved in controlling transcription of a specific gene by binding to a regulatory transcription protein.
- *Oshima*- yeast galactose metabolism.
- **Promoter proximal elements**: regulatory sequences that are located close to the promoter and bind to regulatory proteins.
- Promoter proximal elements have sequences that are unique to specific sets of genes.
- *Tonegawa*- discovered a regulatory sequence within one of the introns that was required for transcription of that gene.
- ^ remarkable because:
    - 1. Regulatory sequence was thousands of bases away from the promoter.
    - 2. It was downstream of the promoter, not upstream.
- **Enhancers**: regulatory sequences that far from the promoter and activate transcription.
- Enhancers occur in all eukaryotes and have several key characteristics
    - Can be 1000+ base pairs away from promoter, can be located in introns or non-transcribed sequences on either prime end of the gene.
    - Most genes have more than one enhancer.
    - Enhancers usually have binding sites for more than one protein.
    - Enhancers can work in both the forward or backward direction.
- **Transcriptional activators**: regulatory proteins that activate transcription.
- **Silencers**: regulatory sequences that inhibit transcription.
- **Repressors**: regulatory proteins that bind to silencers to shut own transcription.
- Silencers and repressors are negative control.
- **Differential gene expression** is a result of the production or activation of specific transcription factors. Eukaryotic genes are turned on when transcription factors bind to enhancers and promoter-proximal elements; the genes are turned off when transcription factors bind to silencers or when chromatin is condensed.
- **Basal transcription factors**: proteins present in all eukaryotic cells that bind to promoters and help initiate transcription.

- **Mediator**: a large complex of proteins that acts as a bridge between regulatory transcription factors, basal transcription factors, and RNA polymerase 2.
- Model of transcription initiation in eukaryotes.
    - 1. Transcriptional activators bind to DNA and recruit chromatin remodeling complexes and HATs.
    - 2. The chromatin remodeling complexes and HATs open a swath of chromatin that includes a promoter, promoter-proximal elements, and enhancers.
    - 3. Other transcriptional activators bind to the newly exposed enhancer and proximal promoter elements; basal transcription factors bind to the promoter and recruit RNA polymerase 2.
    - 4. Mediator connects the transcriptional activators and basal transcription factors that are bound to DNA. This happens through DNA looping. RNA polymerase 2 begins transcription.
- Getting RNA polymerase to initiate transcription requires interactions between many proteins. The result is large macromolecular machine that is positioned at genes start side and is capable of starting transcription.

D. 19.4 Post Transcriptional Control

- 1. Slicing RNAs in various ways
- 2. Modifying lifespan of mRNAs
- 3. Altering transcription initiation rate
- 4. Activating or deactivating proteins after transcription.
- During splicing, gene expression is regulated when selected exons are removed from the primary transcript along with introns. As a result, the same primary RNA transcript can yield more than one kind of mature mRNA that consists of different combinations of exons.
- **Alternative splicing**: splicing the same primary RNA transcript in different ways to produce different mature mRNAs and thus different proteins.
- Alternative splicing is controlled by proteins that bind to RNAs in the nucleus and interact with spliceosomes to influence which sequences are used for splicing.
- Given the extent of alternative splicing, the destination of protein coding genes has been changed to the coding and regulatory sequences that direct the production of one or more related mRNAs and polypeptides.
- **RNA interference**: when a tiny single stranded RNA held by a protein complex binds to a complementary sequence in an mRNA. This unleashes either the destruction of mRNA or a block to its translation.
- **RNA interference**:
    - 1. Transcription of a microgene
    - 2. Precursor micro RNA (miRNA0 formed by initial process of transcription in nucleus.
    - 3. Double stranded miRNA formed when enzyme in cytoplasm trims the RNA hairpin into a short double stranded RNA.
    - 4. Mature miRNA formed when double stranded miRNA binds to RISC protein complex and one strand is degraded.
    - 5. miRNA binds to complementary sequence on target mRNA.
    - 6. RISC cuts mRNA or prevents the mRNA from being translated.