Genetics Notes Chapter 11: Regulation of Gene Expression in Bacteria and Their Viruses

11.1 Gene Regulation

- Bacteria are able to both get organic materials from their environment and synthesize these materials themselves.
- However, it is more efficient to just get the materials from their environment so when a certain organic molecule is available in the environment the bacteria represses genes that produce the enzymes to synthesize this molecule and instead focus their energy on importing the material.
- Bacteria must be able to do two things:
- Recognize their environmental conditions in which they should activate or repress the transcription of relevant genes.
- Switch on or off the transcription of a specific gene or group of genes.
- In prokaryotes, the DNA-protein interaction that determines the start of transcription is the DNA segment called the **promoter** and the protein that binds to this site is **RNA polymerase**.
- DNA segments near the promoter region can also serve as binding sites for regulatory proteins called **activators** and **repressors**.
- In bacteria the binding sites for repressors are called **operators**.
- For some genes an activator is required to bind in order for transcription to begin. Referred to as **positive regulation** because the presence of the protein is required
- For other genes a repressor must be blocked from binding to its target site in order for transcription to occur, this is **negative regulation** because the protein binding is prevented.
- DNA-bound activators often help transcription by tethering RNA polymerase to the promoter so that transcription may begin.
- DNA-bound repressors may physically interfere with the binding of the RNA polymerase to the promoter or may impede the movement of RNA polymerase down the DNA chain.
- Together these and their binding sites function as **genetic switches** that change gene expression in response to environmental changes.
- These regulatory proteins, in order to function must have two states, an active and an inactive state.
- These proteins have two different sites in their structure, one the **DNA-binding domain** is what interacts with the DNA. The other is the **allosteric site** which acts as a switch that either turns the protein to functional or nonfunctional.
- The allosteric site interacts with small molecules called **allosteric effectors** which then change the protein's activity and control their ability to bind or not bind to DNA.
- If the genes encoding multiple proteins constitute a single transcription (i.e. they are either all transcribed or not transcribed) the expression of the gene is said to be **coordinately regulated.**
- An **operon** is a segment of DNA that encodes a multigenic mRNA and its promoter and regulatory region.
- In the **lac system** when lactose binds to the repressor protein's allosteric site it undergoes an allosteric transition, a slight change in the protein's shape, making it no longer have an affinity for the operator. Thus it falls off the DNA and allows for transcription of genes that produce the enzymes necessary to process lactose.
- This relief of repression systems is called **induction.**

- The allosteric molecules that inactivate the repressor leading to gene expression are called **inducers**.

11.2-Discovery of the lac system: Negative Control

- Two scientists, Jacob and Monod, used E. coli's lactose metabolism system to study enzymatic induction: the appearance of an enzyme only in the presence of its substrate.
- When the inducer, lactose, is present the cell creates 1000 times the amount of the enzyme **beta-galactosidase** than it does when lactose is not present.
- This mechanism is very fast as the increase in the enzyme production quickly starts after the introduction of lactose and quickly stops after lactose is no longer produced.
- The induction of beta-galactosidase also occurred concurrently with the induction of the enzymes permease (which transports lactose into the cell) and transacetylase (function unknown).
- Although these enzymes are encoded by three different genes genetic mapping shows that these genes are very closely linked on a chromosome.
- Jacob and Monod induced various mutants of the E. coli bacteria to see how these mutations affected expression. They were able to look at dominance/recessiveness of the mutant-types by creating partial diploids of E. coli using the F' plasmids.
- They found that mutations that inactivate the B-galactosidase gene (gene Z-) and the permease gene (gene Y-) were both recessive to the wild-type Z+ and Y+.
- The scientists discovered two classes of regulatory mutations called **constitutive mutations**. Constitutive mutations caused the lac operon genes to be expressed even in the absence of the inducer (lactose). Essentially the operon was always turned "on".
- The first was the **O^c mutation** which is a **cis-acting mutation**. The O gene does not produce a protein product but is simply a binding site.
- The second mutation the **I gene mutant** is recessive to the I wild-type. This means that one copy of the I gene produces enough protein product (in this case repressor proteins) to maintain cell function (haplosufficient) this gene is **trans-acting**.
- A third I allele was discovered called I^s or the **superrepressor allele**. This allele resulted in the repression of the operon even in the presence of the inducer (i.e. lactose). This allele was dominant to the wild-type and served as evidence that there was a mutation of the repressor's allosteric site in which the lactose could not bind to the allosteric site, therefore the operon was never expressed.
- It was discovered that the operon is encoded by a short sequence of DNA, it has high recognition specificity by the repressor. Even if the operon is just one nucleotide off the repressor will not recognize and bind to it.
- Some of these X, Y, and Z genes are found to be polar. Meaning, they affect the expression of genes downstream from them.

11.3-Catabolite Repression of the lac operon: Positive control

- The first condition for the expression of the lac operon is that (as previously noted) lactose is present as an inducer.
- The second condition however, is that **glucose is NOT present.** This is because the cell gets more energy from breaking down glucose instead of lactose. Therefore it is more efficient to be

breaking down the present glucose rather than building all the enzymes needed for breaking down lower energy sugars such as lactose.

- The control of this second condition is called **catabolite repression.**
- If both glucose and lactose are present in the cell, the X, Y, and Z genes will not be expressed until all of the glucose is already used up because it is inefficient for the cell to produce all the necessary enzymes to break down lactose and other sugars.
- It was discovered that an important molecule **cAMP** appears in high concentrations when there is no glucose in the cell and low concentrations when there is glucose in the cell. It was discovered that the lac operon can only be expressed under conditions where there are high concentrations of cAMP (i.e. when there was no glucose in the cell).
- It was discovered that there is another protein called **CAP** that has the ability to bind with the operon and give RNA polymerase a higher affinity to bind and transcribe the operon.
- CAP cannot bind to the site alone but uses cAMP to bind to its allosteric site which increases the affinity it has with the lac operon thereby activating the lac operon only in the presence of high concentrations of cAMP.
- Overall, the inducer-repressor protein in the lac operon is an **example of negative control.** The protein must be deactivated in order for the operon to be expressed. The cAMP-CAP binding is an **example of positive control** because the complex must bind to the site in order to allow for expression of the operon.

11.4-Dual Negative and Positive Control: The Arabinose Operon

- The genes araA, araB, and araD code for enzymatic proteins that break down the sugar arabinose.
- Transcription begins at the ara I initiation site where an activator protein binds.
- An araC gene (control gene) maps for an activator protein which when bound to arabinose at its allosteric site activates the transcription of the ara operon.
- Additionally, the same cAMP-CAP molecule as the lac operon must bind in order for there to be ara expression.
- In the absence of arabinose, the araC activator protein has a different conformation (shape) it binds to the DNA in two places thereby forming a loop shape that blocks the binding of RNA polymerase, thereby acting as a repressor and stopping the expression of the ara operon.

11.5-Metabolic Pathways and Additional Levels of Regulation: Attenuation

- In bacteria genes that code for enzymes in the same biochemical pathway are often organized into operons and often occur in the order that they are coded for in the DNA.
- In the E. coli **tryptophan operon** one mechanism of repression is the same as that of the lac operon. In the presence of tryptophan that minds to the allosteric site of the repressor protein causing the protein to bind to the DNA and block the initiation of transcription.
- A second method of repression in amino acid biosynthesis operons is **attenuation** which controls the level of transcription by mediating the amount of amino acids and the translation of a "leader peptide".

11.6-Bacteriophage Life Cycles: More Regulators, Complex Operons

- The bacteriophage lambda follows two possible life cycles when it infects a bacterium: