The DNA of different tissues and cell types is the same in a specific organism unlike the RNA and protein content. Gene regulation must therefore operate to produce different amounts of different RNA in different cell types, from the same DNA.

**Learning Objective**

After interacting with this Learning Object, the learner will be able to,
- Define basics of gene regulation.
- Describe gene regulation in Bacteria.
- Describe gene regulation in Eukaryotes.
Basics of gene regulation

In prokaryotes, transcription by RNA polymerase can take place with the help of an activator protein. However, in the presence of a repressor molecule, the binding site for RNA polymerase is inaccessible due to which transcription does not occur. In the ground state, the repressor does not remain bound because of which the gene is turned “on”.
Basics of gene regulation

The regulatory site of DNA in eukaryotes remains inaccessible for binding by transcription machinery due to which gene expression remains turned off in the ground state. In the active state, however, the DNA forms a loop thereby bringing together the promoter proximal element and enhancer. Interactions of these two elements with RNA polymerase successfully activates expression of the gene.
An inducible system is ‘off’ in its ground state and must be turned on by an effector molecule, which is known as the **inducer**. In the negative regulation mechanism, the inducer binds to repressor and prevents it from binding to the operator region. This allows RNA polymerase to proceed with transcription by binding to the promoter. In positive regulation mechanism however, the inducer binds to the inactive activator to produce the active activator molecule which in turn facilitates binding of RNA polymerase to the promoter to turn on expression.

Gene regulation in prokaryotes
Gene regulation in prokaryotes

The ground state in case of a repressible system is ‘on’. It has to be turned off by an effector molecule, which is known as the **co-repressor**. In case of negative regulation mechanism, the co-repressor binds to the inactive repressor molecule and activates it, thereby preventing gene expression. In positive regulation, the co-repressor binds to the activator molecule and prevents its binding to the promoter region, thereby turning off gene expression.
Gene regulation in prokaryotes

The lac operon consists of a group of genes that are responsible for transport and metabolism of lactose sugar in certain bacteria like *E. coli*. This operon is under negative regulation by the LacI repressor protein. In absence of the inducer, the tetrameric repressor binds to the operator region, thereby preventing transcription by RNA Polymerase. In presence of the inducer, the inducer binds to the repressor protein which then prevents it from binding to the operator and therefore allows gene expression.
Gene regulation in prokaryotes

*Lac* operon also undergoes positive regulation by means of the cAMP-CAP system. Glucose is the preferred energy source for bacteria and if both glucose and lactose are present, β-galactosidase enzyme which metabolizes lactose is not synthesized. High glucose levels prevent synthesis of cAMP which is essential for binding to the catabolite activator protein. This protein facilitates transcription of the *lac* operon. When glucose levels are low, cAMP is produced which binds to the CAP, which in turn binds to a distal part of the promoter region and facilitates transcriptions.
Gene regulation in eukaryotes

Eukaryotic gene regulation is a complex process that can be regulated at various levels starting from the gene transcription to the post-translational modification of the protein to form the active, functional molecule. These levels include transcription, post-transcriptional modification, translation, post-translational modification and protein transport, the most common of which is the transcriptional level.
Gene regulation in eukaryotes

Chromatin remodelling, which provides accessibility to the gene, is a prerequisite for gene expression and is one of the points of regulation at the transcriptional level. Remodelling involves acetylation of histone proteins and demethylation of DNA which are carried out by various enzymes. Several regulatory proteins are also involved in the transcription process after chromatin remodelling, which serve as important points of regulation.
Gene regulation in eukaryotes

mRNA synthesized from DNA by transcription undergoes several post-transcriptional modifications to form the mature mRNA which then undergoes translation to form proteins. These modifications are also under regulatory control to moderate the amount of mRNA produced based on the requirement. Addition of a poly (A) tail promotes export of the mRNA from the nucleus and protects the mRNA from degradation. mRNA that does not have these modifications is usually very unstable and will be degraded.
Gene regulation in eukaryotes

In eukaryotes, the mRNA does not get translated until it receives the appropriate signal, thereby serving as another point of control. Before receiving the signal, enzymes required for the process will not be present and will get synthesized only after the signal is obtained.
Gene regulation in eukaryotes

Proteins undergo several post-translational modifications such as phosphorylation, acetylation, methylation, glycosylation etc. These processes are essential for formation of the active, functional protein. Regulation at the post-translational level can lead to degradation of the non-functional protein molecules.
Basics of Gene regulation

1. Gene regulation: The process by which the synthesis of a gene’s mRNA transcript and its corresponding protein product is controlled or regulated by various signal molecules is termed as gene regulation. In this process, a cell determines which genes to express and when to express them. Regulation of processes is essential to ensure that no wastage of energy or cellular materials takes place.

2. Single celled organisms: Organisms that have only one cell containing all the organelles and genetic material within one common compartment are known as single celled organisms. The bacterial genome has 4000 genes of which only a fraction of them are expressed at any given time. Moreover, requirements for gene products vary with time such that some products are required in large amounts while others in smaller quantities.

3. Multi-cellular organisms: Gene regulation is a more complex process in eukaryotic and multi-cellular organisms that contain more number of cell organelles, each having complex processes taking place in them. The human genome contains around 35,000 genes, out of which only a fraction of them are expressed in a cell at any given time. Gene expression varies in different cell types even though their copy of the genome is identical. Certain genes, known as housekeeping genes, are expressed in all cells while others are specific only to certain cell types. For example, the gene for glucagon hormone is expressed only in pancreatic cells while antibody synthesis genes are continuously expressed in plasma cells.
Basics of Gene regulation

4. **Transcription**: Transcription is the process by which information from a double stranded DNA molecule is converted into a chemically related single stranded RNA molecule by making use of one strand as the template. Transcription which takes place in the eukaryotic nucleus is separated in space and time from translation taking place in the cytoplasm.

5. **Translation**: A process by which the mRNA sequence is read in the form of three letter codes known as codons to incorporate the corresponding amino acids in the growing polypeptide chain with the active involvement of rRNA, tRNA and several other enzymes.

6. **Different levels of gene regulation**: Gene regulation can be carried out at several levels starting from the synthesis of the mRNA transcript till the degradation of its corresponding protein product. The various stages include:
   a. Synthesis of the primary RNA transcript (transcription)
   b. Post-transcriptional modification of mRNA
   c. Messenger RNA (mRNA) degradation
   d. Protein synthesis (translation)
   e. Posttranslational modification of proteins
   f. Protein targeting and transport
   g. Protein degradation.
Basics of Gene regulation

7. **Active state:** The state in which the gene is turned “on” and synthesizes its corresponding mRNA and protein.

8. **Repressed state:** The state in which the gene is turned “off” and no transcription occurs. This could be due to binding of a repressor molecule to the gene.
1. **Operon**: A functioning unit of genomic material that is made up of a cluster of functionally related genes that are under the control of a single regulatory element. Operon arrangements are a commonly observed mechanism of gene regulation in prokaryotes and can be either inducible or repressible.

2. **Lac operon**: The first system of gene regulation that was understood in *E. coli*, worked out by Francois Jacob and Jacques Monod in 1962. The lac operon is negatively controlled by the lacI repressor and positively regulated by catabolic activator protein (CAP).

3. **Operator**: Operators are regions of DNA that are around 15 nucleotides long and are generally located near a promoter element such that they control the access of RNA polymerase to this region.

4. **Promoter**: The region of DNA to which RNA Polymerase binds and starts the process of transcription.

5. **Activator**: A molecule that enhances the interaction between RNA polymerase and a particular promoter region, thereby facilitating expression of the gene.

6. **Repressor**: A protein that binds to a regulatory region such as an operator, adjacent to a gene and thereby prevents its transcription by impeding the binding of RNA polymerase.

7. **Effectors**: Effectors are molecules that affect the binding of activators and repressors to the operator region of DNA. These can either be inducers, in case of inducible systems, or a co-repressors if it is a repressible system.
8. **Inducible system:** An inducible system is originally ‘off’ in its ground state and must be turned on by an effector molecule, which is known as the **inducer**. In the negative regulation mechanism, the inducer binds to repressor and prevents it from binding to the operator region. This allows RNA polymerase to proceed with transcription by binding to the promoter. In positive regulation mechanism however, the inducer binds to the inactive activator to produce the active activator molecule which in turn facilitates binding of RNA polymerase to the promoter to turn on expression.

9. **Repressible system:** The ground state in case of a repressible system is ‘on’ and it has to be turned off by an effector molecule, which is known as the **co-repressor**. In case of negative regulation mechanism, the co-repressor binds to the inactive repressor molecule and activates it, thereby preventing gene expression. In positive regulation, on the other hand, the co-repressor binds to the activator molecule and prevents its binding to the promoter region, thereby turning off gene expression.
1. **Gene expression:** The process of transfer of genetic information from the nucleotide sequence level in a gene to the level of amino acid sequence in a protein or the nucleotide sequence of mRNA is known as gene expression.

2. **Eukaryotic regulation:** Eukaryotic cells have larger and more complex multimeric regulatory proteins when compared to bacterial cells. The process of regulation is therefore, also more complex and can be achieved either by altering the rate of transcription, the stability of mRNA molecule or through regulation at a translational level. Regulatory elements that control these processes may be tissue specific, thereby activating or deactivating genes only in one kind of tissue.

3. **Chromatin:** DNA that is packaged with basic proteins known as histones form a structure known as chromatin in eukaryotes. This chromatin structure helps in restricting access to eukaryotic promoter sites. For gene expression to take place, remodelling of the chromatin must occur wherein, acetylation of histone proteins and demethylation of DNA occur, which then favours transcription.

4. **Promoter:** The region of DNA to which RNA Polymerase binds and starts the process of transcription. The promoter in eukaryotes contains a sequence of 7 bases known as the TATA box, which is bound by a large number of proteins including the TATA-binding protein (TBP), and various transcription factors.
Gene regulation in Eukaryotes

5. **Exons**: The regions of mRNA that code for specific proteins or entire protein products upon translation are known as exons. They are often discontinuous with intervening nucleic acid sequences being present between them.

6. **Introns**: The *intragenic* sequences, sometimes considered as “junk”, that are present in the pre-mRNA but do not get translated into proteins are known as introns. These are removed during the process of RNA splicing.

7. **Enhancers**: Enhancers are sequences of DNA to which regulatory proteins can bind. Most of these are located outside of the promoter region. Binding of transcription factors to enhancers is associated with an increase in the rate of transcription.

8. **Silencers**: Silencers are control regions of DNA, onto which transcription factors bind in order to decrease the rate of transcription.

9. **RNA splicing/post-transcriptional modification**: It is the process by which exons are spliced or cut out from the pre-mRNA molecule to give a coding, mature mRNA sequence, which is then translated into protein. This is another point at which gene regulation commonly occurs. Splicing can take place such that the exons are re-joined in different combinations, in a process known as alternative splicing. This allows a variety of different polypeptides to be translated from a single gene.
10. **RNA Transport:** The process by which only fully processed and mature mRNA is allowed to leave the nucleus in order to be translated into protein. Any defective mRNA will be degraded within the nucleus itself.

11. **Positive regulation:** Although eukaryotic cells exhibit both positive and negative regulatory mechanisms, the positive mechanisms have been found to predominate in all systems characterized so far. The transcriptional ground state is therefore restrictive or silenced, and virtually every eukaryotic gene requires activation before it can be transcribed.
Negative regulation of the lac operon is brought about by:

- cAMP
- lacI
- CAP
- None of the above

Congratulations, you have chosen the correct answer.
Gene regulation is most commonly implemented at which of the following stages?

- translation
- transcription
- post-translational
- protein transport

Congratulations, you have chosen the correct answer.
Which of the following is the effector molecule for the repressible system?

Activator
Co-repressor
Inducer
Repressor

Congratulations, you have chosen the correct answer.
What is the binding interaction that takes place during negative regulation in an inducible system?

- Inducer-repressor (Correct)
- Inducer-inactive activator
- Co-repressor - inactive
- Co-repressor - activator

Congratulations, you have chosen the correct answer.
The binding site for the effector molecule is?

- Promoter
- Enhancer
- Operator
- None of the above

Congratulations, you have chosen the correct answer.
Books:

2) An introduction to Genetics by Griffiths, 8th edition.