NOW we will talk about the second level TRANSCRIPTIONAL level it is the most important level?? because once the cell has decided to copy some gene it will think about this level that it will copy million and a half copy and make a processing to them within a period of time then post transcription modification then translation then post translation modification except if there is an emergency like the cell discover that it need more or less of this protein or deferent types so it is possible to as the post transcriptional level or translational level.

Example :- if someone take a drug then the liver cells decided the amount of RNA needed to produce proteins to detoxification of the drug suddenly after the transcription and translation finish the patient take a diuretic then the drug excretion will increase so before that 10,000,000 RNA were reached to the cytoplasm now we don't want all of them to reach and if they reach the cell doesn't translate all of them if it translate all it doesn't modification all of them on the post translational level.

What control the transcriptional level?! we have two cis acting elements and two trans acting elements Cis is from the DNA itself , Trans is protein bind to the element),so when we talk about origin of replication which has splitting of the two strands it is CIS ,,,and when we talk about the helicase it is TRANS.

In the transcription the cis elements are the Promoters and Enhancers, the promoter present before the gene exactly but the enhancers have no specific position, Both of them have proteins bind to them called Transcriptional Factors which are the TRANS elements, these factors brings the RNA polymerase which can't identify a promoter without the factors so they are important.

The promoter is consensus element at the 5' region (consensus position and sequence) e.g: TATA box is the most important sequence of promoters (not the only one) is located at the position from -10 to -20 at all genes, GC box -30 -110 and so on, but The enhancer may be close or far, Both of them have transcriptional factors and the question here how do they communicate????Actually the DNA will BEND to bring them closer but that is not enough, they need mediators proteins between them (Transcription factors are called general or basal on the promoter, since it's the same for all genes >> specific on the enhancer, as it varies for each gene).

The basal transcriptional factors have low efficiency to stimulate the RNA polymerase but when the enhancer work with it the efficiency will increase (if the promoter makes 10 copies, with the enhancer they will get 10 million copies), that is why it called enhancer.

Remember TATA box is not the only sequence of promoters, it is one of the core promoter region (which is a region of promoter), TATA binding protein is the protein binding on the TATA box, to that protein the transcriptional factors bind (the general factors), why??? Because not all the proteins can read the DNA and identify the promoter, but the TATA binding protein can do it , basal

cannot and the RNA polymerase cannot, however, the efficiency of the TATA binding proteins and proteins bound to it is low in initiating transcription, unless it gets a massage from the enhancer by the mediators.

There are different names in the books e.g Trans element has many names like specific or Trans (means activating from distance) or activators or hormone receptor (because they're activated by hormones (cortisol, T3…ETC) the most used name). Mediator proteins are called Co-activators

So many promoters with many enhancers communicate through mediators>>bring the RNA polymerase to make a million copies, and when it finishes the transcription factors will dissociate, so they told it when to start but what told it to stop?? The termination signal, where the transcription factors dissociate.

Note: - the students mix between transcription and translation start and stop, in transcription there is a promoter and terminator, in translation start and stop codons.

Q: do all the genes have promoter and enhancer???

A: yes and the enhancer give the specificity.

***(In slide 199 + 201): here we will give specific example for specific transcription factor or tans-activator or activator or hormone receptor and here it's the glucocorticoid hormone receptor. Gr: it's the glucocorticoid receptor and the hormone is cortisol now the Gr in this situation it's expressed but it's found in the cytoplasm as a transcription factor, it functions in the nucleus but it can't enter the nucleus due to HSP protein (heat shock protein). When the body secretes cortisol it will bind to Gr and the HSP will dissociate because cortisol has high affinity so now the Gr will be switched from the inactive form to the active form, and immediately it will enter the nucleus and functions as a transcription factor on its enhancer GRE (glucocorticoids respond element)**

When the Gr was bound to the HSP it couldn't enter the nucleus but after its binding to the cortisol it enters the nucleus, why?? All proteins are produced in the cytoplasm there is a group of proteins called nuclear proteins which function in the nucleus not in the cytoplasm so when they are produced in the cytoplasm these nuclear protein will be transferred to the nucleus, how?? By proteins (shuttle proteins and transport proteins) and they can differentiate between nuclear and cytoplasmic proteins by a polypeptide signal (nuclear localizing signal 'NLS') presented on nuclear proteins. In Gr the HSP is a huge protein masking the 'NLS', on the other hand the cortisol is small so when it binds to the Gr the 'NLS' will be exposed and it can enter the nucleus . When the Gr enters the nucleus it will go to its place (which is any gene that carry Gr enhancer 'GRE') then co-activators and mediators come and activate the promoter and the basal transcription factors and transcription will occur.

***We can see that Gr is like many other specific transcription factors or hormones receptors or trans-activators they work in dimmers and here we call it a homo dimmer (two subunits of Gr work together).**

***Some hormones have less effect than other hormones why?? Apparently it depends on how many genes are activated by the receptor of that hormone. For example: hormone is secreted in blood enters all the cells, cells that have receptors for that hormone will respond (the receptor is a transcription factor that activates some genes by the enhancers for this factor) it could be one gene or twenty or more and this will determine the effect. The receptor for cortisol will bind to a huge number of genes that are important for metabolic activity of the cells and that's why it's secreted in small doses and short periods.**

***Another transcription factor which is another hormone receptor it's called the thyroid receptor (slide 200+202) and its enhancers is TRE (thyroid response element). The thyroid receptor present in the nucleus so it will go directly to its enhancer but when the thyroid receptor binds without thyroid hormone it will not attract activators but it will attract repressors, but when T3 binds to the receptor its shape will change and it will attract co-activators and the transcription complex will receive message and it will bring the polymerase for copying.**

In this example how some co-repressors and activators work? In slide 202 the repressors have histones deacetylase activity (HDAC domains) and in the activators there is histone acetylase activity (HCA) what does that mean?

In the first structure here the thyroid receptor is alone(without T3 hormone) and it is attached to co-repressor that has HDAC which remove any acetyl group from histones near DNA and the positive charge will appear and attract the DNA and there will be no transcription . When the thyroid hormone T3 binds to the TR and brings co-activator that has HAC they make acetylation for the near histones and the positive charge disappears and transcription increased.

***TR here works as a hetero dimmer and its partner isn't another TR but it's RXR (retinoid x receptor)**

Most of transcription factors that are hormone receptors they work in dimmers either homo dimmers or hetero and most of hetero that don't have homo dimmer the partner is RXR (RXR is a very good target for some good cancers therapeutics)

***slide203: Remember that some of transcription factors has the ability to catch the DNA and read it , these have specific structural motives or domains , some of these motives are Zinc fingers ,B-Zipper proteins , Helix-turn-helix and Helix loop helix. (Just to know without details)**

Transcription factors are responsible for controlling our genes, but who are responsible for controlling transcription factors?

1) Ligands, most of TFs work as receptors if we remove the hormone TFs will not work.

2) Or by partners with RXR remove the RXR TFS will not work.

3) Most of TFs are phosphoproteins if we add to them phosphates they will work remove it the will not work and some of them are the opposite if we put phosphate they work if we don't they will not work.

4) There is another way for controlling TFs; TFs is protein and has gene and this gene has promoter attached to another TF so if I control the second TF I will prevent the production of the first TF and the second has gene and it has promoter and it attached to another TF and so I will go back until I reach the top of transcription factor which are called the master TF which are responsible for activation all of TFs which means they activate all the genes in our body.

Now, there are companies that give you transcription factors like SOX2, NANO, OCT4 that let you makes zygotic cells from differentiated ones... One more example is -- if you have a defective valve, doctors can take a biopsy of your buccal smear and adding transcription factors to produce embryonic cell then adding some substances giving you heart valve. This process is good b/c there is no rejection, recipient, donor, or transplantation. This process is known as Induced Pluripotent Stem Cells (IPSCs). This is due to master transcriptional factors.

MULTIPLE REGULATORS OF PROMOTERS.

Note that enhancers are specific.

How many enhancers can be recognized by glucocortisone when you take it? 2 or 3. But how glucocortisone activates 50 genes? When cortisone binds to its receptor, there will be response by their enhancers A & B. A and B will give protein which is by-self is a transcription factor. This transcription factor will activate genes E, F, and G. Gene G may produce other proteins that activate other lines of genes. This process is known as "waves of gene amplification". At the end, the last protein of the last wave makes a negative feedback on the first gene. All the activation depends on the binding of cortisone to its receptor.

Note that promoter or enhancer region element has multiple loci like cortisone region element, thyroid response element and others. It depends on which ligand binds to give different response. Sometimes, there is concordance between two elements to produce specific response.

POST TRANSCRIPTIONAL LEVEL:

There are two qualitative examples:

1. **Alternative splicing and alternative polyadenylation.**
2. **RNA editing.**

Alternative splicing:

The same gene giving the same mRNA, but eventually we get different proteins in cells of different tissues, as the mRNA is spliced differently.

For ex: in thyroid certain introns are removed to give Calcitonin, while in the brain different set of introns are removed to give another protein CGRP.

Also, alternative polyadenylation takes place to give the different proteins.

RNA EDITING:

DNA → RNA → PROTEIN

RNA which is related to Apo-protein B gene in the liver cells gives a protein "its length is 4563 a.a".

RNA which is related to Apo-protein B gene in the intestine gives a protein "its length is 2152 a.a". What is the difference between them is the changing in the sequence of the codon from CAA to UAA. UAA is a stop codon. When it gets translation, instead of getting protein like in liver cell, intestine cell synthesizes truncated short protein b/c it used the stop codon. This mechanism of changing from codon to non-codon is unknown.

Thank u

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