Regulation of gene expression

❖ How do we regulate our genes?

- Each cell in our body has the same number of genes so what's make cell different from another is the expression of genes "**expression profile**"
- Expression >> when a gene is expressed that means :

DNA>>RNA>>Protein

- expression profile ⟶ which gene is being expressed in that cell at that given moment
- That means it differs from a cell to another depending on its specialty

And within the cell according to its condition

- So the cell is continuously switching ON AND OFF expressing it's genes

But there are genes that are always on and others always OFF

- For example
- The globulin gene is always on the RBC's
- Insulin genes are always on in the Beta cells
- The genes that are ALWAYS ON are called the "**housekeeping genes**"

They are essential in the life of the cell

- The genes are not just turned on or off but there is a level of that " level

Of expression"

- The expression level is very important because if the level of expression for some genes changed it may cause some problems
- For example P53 gene if it was expressed 10% more it may cause apoptosis and if it decreased by 10% some mutations may happen

➢ Cells are able to switch on/off certain genes when needed according to:

a. Development period: (e.g.. Fertilized egg-------adult human)

Through the cell development the expression differs

b. Differentiation stage: (e.g.. Stem cell--------Lymphocyte)

Cell differentiation inside the full body

c. Environmental changes (e.g.. UV and melanin production)

In differentiated cells

For example :

- If the skin cell was exposed to the sun it will produce more melanin
- The pancreas cells >> depending on the diet they change the insulin

secretion rate

- The liver cells >> when you take a medicine the detoxification genes will be expressed

➢ Between the prokaryotes and eukaryotes gene expression there is a lot in common but we'll start talking about the differences

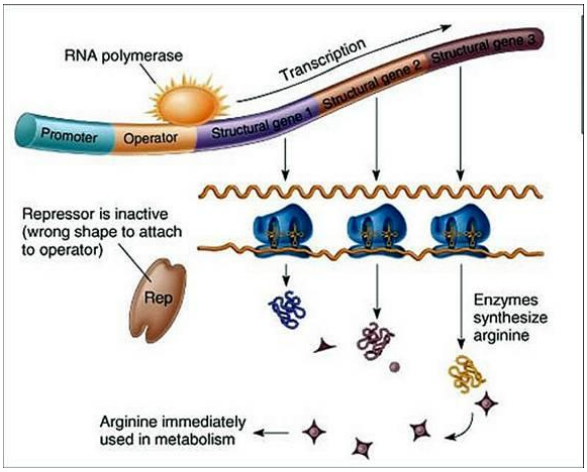
Prokaryotes differ by having:

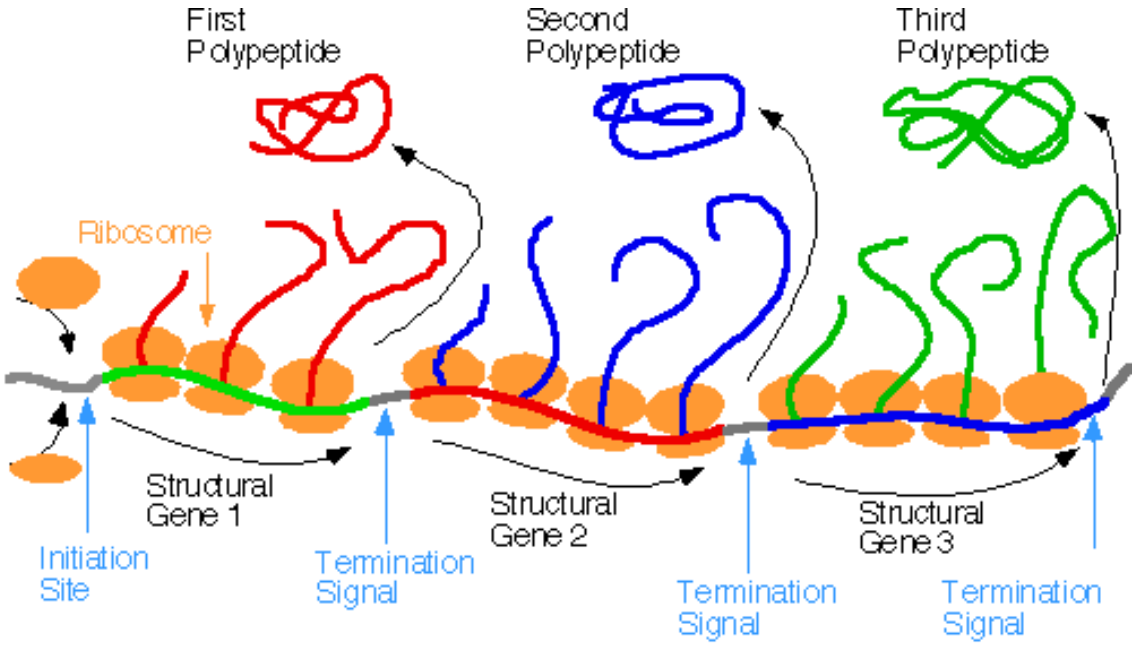
1. Operons
2. Operator
3. Inducer
4. Repressor
5. Co- Repressor
6. Attenuation of transcription
7. Sigma factors

❖ **<u>Operons</u>** :

- A group of genes under the control of the same promoter
- Us as humans each gene is controlled with a promoter >> 30000 genes with 30000 promoters
- But prokaryotes have group of related genes that are controlled with one promoter

  Since the products of these genes work together on the same thing so they need them as a pack
- The expression of the operons will produce a single **<u>polycistronic mRNA</u>** and it contains a multiple sets of start and stop codons





In slide 165

- We have 3 start and stop sets
- 3 reading frames
- Each part is translated by ribosome's giving us different polypeptide but these polypeptides have related function

✓ The first operon to be discovered is the **lac operon** (lactose metabolism genes)

- The bacteria usually produce its energy from glucose but when it runs out of glucose it will use the lactose
- So it will need protein channels to get the lactose in then it needs enzymes to break down the lactose to glucose and galactose after that it needs an enzyme to turn the galactose to glucose

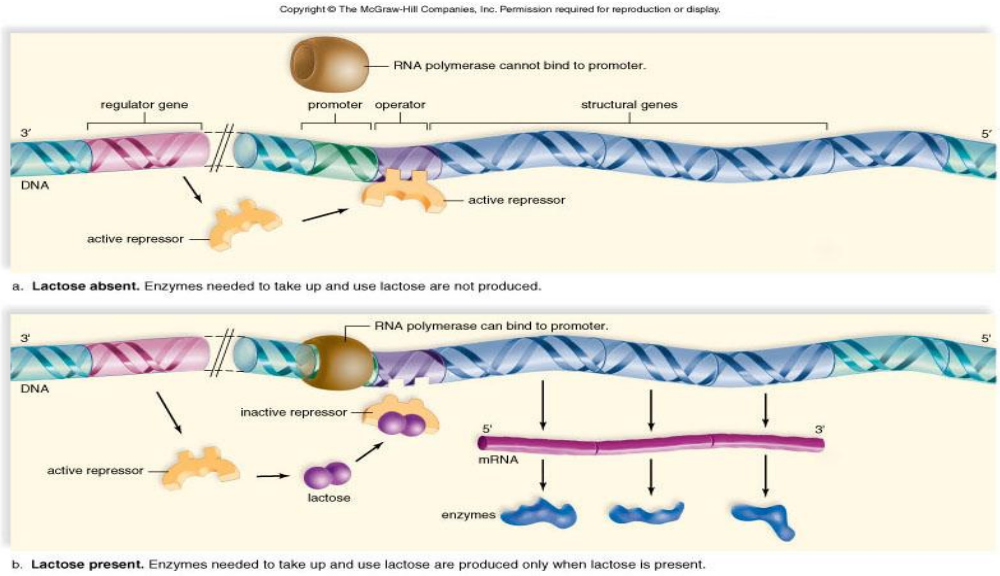
- We need these enzymes at the same time so the genes that are responsible for making these enzymes are found with a single promoter therefore all the genes are expressed at the same time

- The cell dose not activate the lac operon just by the presence of the lactose it should be running out of glucose

- How does the cell know that there is no glucose ?

By secondary metabolites

glucose 》》》 cAMP 》》》 cAMP + CRP 》》》 induction of the lac operon



❖ Operator :

- sequence that is overlapping with the promoter
- It's like an extra check point
- It's a sequence that's downstream of the promoter that can attach to a protein called the **repressor**
  - So if the promoter bind to RNA polymerase the operator will stop it from working by binding to the repressor
  - The **Inducer** remove the repressor

✓ In some genes usually they are repressed and we bring the inducer to remove it but in other genes there is no repressor since we always need the gene but if we need to stop expressing it we put a repressor by the help of a **Co-Repressor** .

❖ Attenuation of transcription

- attenuation >weakening in force or intensity
- Reduce the transcription
  - Since there is no nucleus in prokaryotes the transcription and translation can occur at the same time (no segregation as no compartmenting)

➢ So ribosome should move smoothly or it may remove the polymerase and the transcription will not finish

- By this the cell control the expression

- ✓ For example **trp(tryptophan)operon**
- The cell produce it when it runs out
- The polymerase start in transcription of the trp operon but the first 5-6 codons are anti trp codons so if there is trp in the cell it will bind with the ribosoms and it will make them move faster so the polymerase will detach and there will be no production of trp
- But on the other hand if there was no trp in the cell the ribosomes will move slowly and the polymerase will continue to work normally

❖ Sigma factors

- RNA polymerase in prokaryotes is different from RNA polymerase in eukaryotes
- The RNA polymerase in prokaryotes are blind
- They can't see the promoter
- They need a protein "sigma factor " to transfer it to the promoter
- But in humans the RNA polymerase can go by itself and bind with the promoter

- And this is really helpful in making antibiotics since we focus on the differences between us and the bacteria so we will be able to make a drug that only attack the bacteria and the sigma factor would be a fantastic target .

Regulation of gene expression in eukaryotes (chapter 16)

- It's a little bit complicated, why?
    1. The presence of the nucleus, which means segregation between transcription and translation:
       m RNA is produced, processed, then it exits the nucleus, translated into protein in the cytoplasm by ribosomes, then is being folded.
    2. Our DNA is NOT alone:
       It's wound around histones, so when transcription is needed, DNA is being unwound, transcribed and rewound again.
    3. Our cells are diploid, in contrast with bacterial cells which are haploid:
       We have two copies of each gene; when one gene is turned on, the other gene must be turned on. But in bacteria, there's only one gene is turned on.
    4. We have much more DNA:
       For example: E.coli has 3,000 genes, but we have 30,000 genes.
    5. The presence of introns and non-coding regions:
       As if the bacteria has 3,000 sequential short stories, but we have 30,000 sequential short stories, and each one has a lot of parentheses.
    6. Each gene is controlled by a single promoter:
       There's no operons , nor group regulation. And that helps in expression of the gene in high speed.
- What counts in the end, is the quantity and quality of the **functional protein**.
- How can we get functional proteins? By regulation at different levels:
    - DNA and chromosomal level (the gene status).
    - Transcriptional level (the promoter activity, the number of m RNA produced).
    - Posttranscriptional level (m RNA processing).
    - Translation level (the ribosomes activity).
    - Posttranslational level (modification of the polypeptide: like methylation, phosphorylation, cleavage ,or become quaternary protein)

- DNA and chromosomal level:
    - Availability of gene expression:
      Is this gene accessible for transcription or not.
    - There are 5 ways in this level, the first two ways happens in all the cells of our body at DNA level, the others happens in special cells

1. Chromatin remodeling:
    - What is the difference between chromatin and chromosome?
      The same ingredients: DNA around histones, but

	Form	In cell cycle
Chromatin	Random	Almost all the

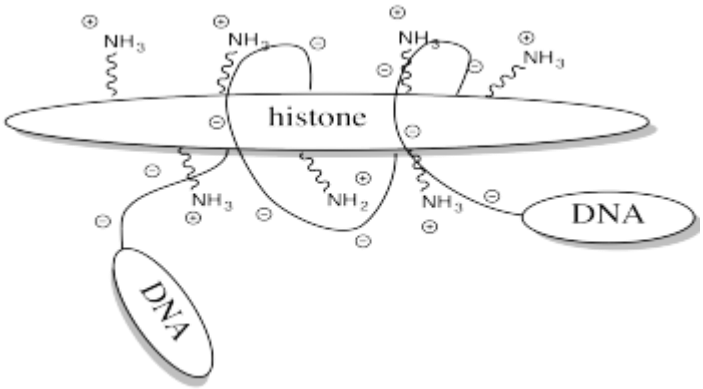
		time (interphase)
Chromosome	Organized	At metaphase

- What is the meaning of chromatin remodeling?
  The changing between <u>heterochromatin</u> (condensed chromatin) and <u>euchromatin</u> (diffused chromatin).

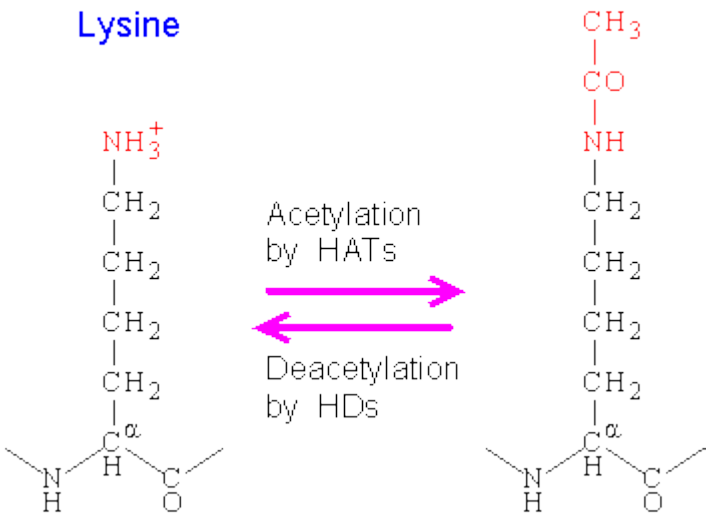
  > Note: The basic unit in DNA packaging is the **Nucleusome**, which is parts of chromosomes wound around histone core tightly or loosely.

  This change is reversible in all cells **EXCEPT FOR** the <u>RBCs</u>(During RBC maturation, it loses a lot of its genome. Any part of its genome does not need it, it packages it, and finally it deletes these parts –not only packaging, because at the end the RBC doesn't need any gene except for the *Globin gene).*
- There are two ways to perform chromatin remodeling:

1. <u>Histone acetylation and deacetylation</u>:
   - It's more used (99%)
   - How does the DNA wound around histones?
     By electrostatic interaction, -DNA is negatively charged
     -Histones are positively charged } Why?
     Because the DNA contains **phosphate group**, which is negative, and histones contain **Lysine** amino acid, which is positively charged.



   - So, How can we release the DNA of the histones?
     By masking the positive charge on histones (covering it) by Histone acetylase using acetyl group.

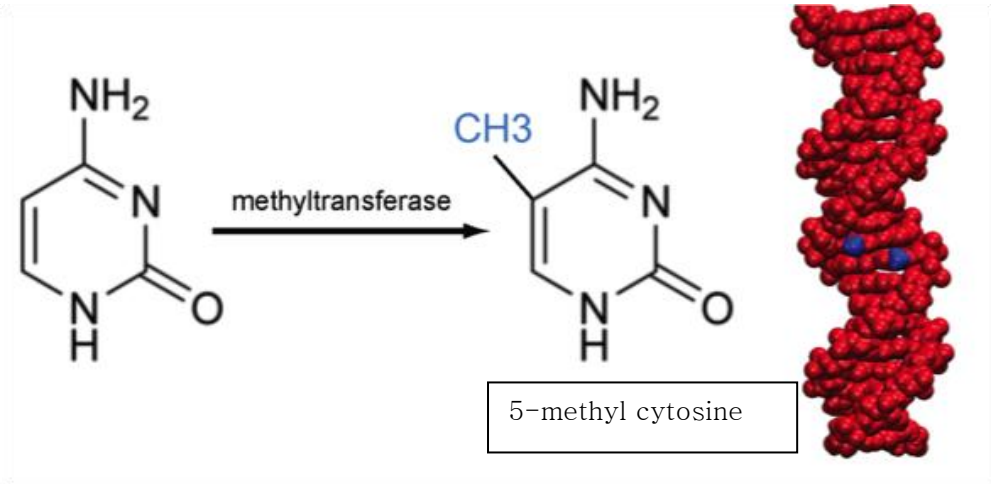


- What removes the acetyl group?
  Histone Deacetylase.

2. ATP-driven way:
   - Less used method (1%)

2. DNA methylation:

- What is DNA methylation?
  Addition of methyl group on carbon number 5 of cytosine residue on the gene promoter (since it's the most nucleotide repeated in promoter region) to prevent transcription of that gene by RNA polymerase, it's like camouflaging the promoter.



- Where would you expect lower levels of methylation of *Globin gene* promoter, in erythroid cell or nonerythroid cells?
  Low levels of methylation means that the gen is **ACTIVE**, so the answer is: In erythroid sells.
  For example: In skin cell, you'll find that the *Globin gene* is heavily methylated because it does not need globin.

Note: DNA methylation in males differ from that of females, Details:
On chromosome 15 there is a region that has 3 genes, in females gene number 2 is methylated, but in males gene number 1 is methylated , according to this fact:
If an individual inherits a deletion in chromosome 15 involving the three genes, depending on from whom he inherited that deletion mutation, he will get a completely different syndromes.
If the deletion occurs in paternal chromosome=loss of gene 1 methylation: Prader-Willi symdrome If the deletion occurs in maternal chromosome=loss of gene 2 methylation: Angleman syndrome

3. Gene rearrangements:

- Specific for B-cells (Antibody-producing cells)
- Immature B-cell does not give all the genes for its future mature cell. Instead, it gives the mature cell only few genes that helps it to produce only one type of antibodies.

4. Gene deletion:

- Specific for RBCs.
- Any nonerythroid cell, when it doesn't need any gene, it turns it off (like chromatin condensation or methylation .. etc), but when RBC cell doesn't need a gene, it deletes it, cuts it and breaks it down, because at the end, this RBC will provide an extra space for hemoglobin.
- This occurs during maturation into fully active RBC.

5. Gene amplification:

- It's a **pathologic** state, happens in cancer cells.
- In normal cell: When the cell wants to over express a gene, it's limited by **two copies** only of genes, so it increases the number of m RNA, and from that we get maximum amount of proteins.
- In cancer cell: It activates oncogenes that helps in cell growth and division.
  (in normal situation, the promoter of this gene is very weak)
  What happens in cancer cell, it transcribes this gene at DNA level (they are called: **Double minutes**), splice them and integrate them on the same chromosome or another one. So, instead of having 2 weak copies of this oncogenes, cancer cell will have 400 weak copies, even they are weak, the cell will divide very quickly.

- The most famous example is : **HER2/NEU (**also called: **erbB-2** , or **EGFR**: Epidermal Growth Factor Receptor**)**
  - It's an oncogene on long arm of chromosome 17, in normal situation: it's under control, in cancer cell: there are so many copies of this gene, as a result a huge number of m RNA and then lots and lots of proteins are produced.
  - This protein is cell receptor on the cell of breast tissue for a growth factor, *Normally.. one receptor .. weak signal for growth

*Cancer cell.. ten thousands of receptor .. strong signal (amplification of the signal) even if the ligand in low concentration… Breast cancer

- 40% of breast cancer is because of gene amplification of *HER2 gene*.
- Nowadays, there is drug that blocks the receptor : Herceptin
- Cancer drugs (Smart drugs) are formed according to protein level that blocks the amplificated protein

Sorry for any mistake ☺

GOOD LUCK ^_^