Hello students, today we will be conducting a e-learning session on the topic gene expression and regulation. In this particular topic of gene expression and regulation, today's module is positive and negative regulation of the lactose operon. in this particular module we will be studying about the structure of the lactose operon. The negative regulation of the lactose operon, and positive regulation of the lactose operon. What is an operon? An operon is a group of genes with a single promoter moiety, and this group of genes is entirely regulated by this single promoter sequence which is present.

This particular operon that is the lactose operon is found primarily in E. Coli,and it is also found in a lot of other bacteria.

The structural genes of the Lac Operon are responsible for degradation of the carbohydrate lactose. As you all know,lactose is made up of two subunits, that is, glucose and galactose. Given an option, the cell would primarily utilize glucose because it is easier to utilize and it generates a lot of ATP. If we take a look at the structure of the lactose operon,we can see that it is made up of structural genes, and it is also made up of regulatory sequences.

The regulatory sequences are the promoter region and the operator

region and the structural genes are the lac Z, lac Y and lac A genes.

lac Z codes for the enzyme beta galactosidase, which

is the one which is responsible for breaking down lactose into

glucose and galactose.

The second structural gene that is the lac Y gene is

responsible for producing the enzyme permease and this is

responsible for uptake of lactose into the cell. The

lac A gene is responsible for

production of the enzyme transacetylase, and this also helps

in breakdown of that lactose.

In addition to this, the regulatory sequences are the

promoter region and the operator

region. The operator region is present in between the

promoter region and the first structural gene.

The lactose operon is also influenced by two other

elements. The first gene is theLac I gene, the Lac I gene is

the gene which is responsible for production or generation of

their repressor protein and this particular Lac I gene is

regulated by the promoter which is the promoter of the repressor gene.lactose Operon is a catabolic operon responsible for breakdown of lactose, and by default the lactose operon is always turned off.

Which means that the repressor protein is always being produced constitutively and the repressor protein will go and it will attach to the operator molecule moiety an as a result, when the repressor protein is attached to the repressor moiety, the RNA polymerase cannot carry out the process of transcription. Where does the RNA polymerase bind ?The RNA polymerase binds to the promoter region and as you can see the. Promoter Region is present upstream of the operator region and therefore, when the repressor molecule protein is bound to the operator region,the RNA polymerase cannot cross it and go on to transcribe the structural genes, so no transcription occurs because repressor protein blocks RNA polymerase. How does this happen? The repressor protein is a tetramer, it binds to two sites in the operator molecule. And in addition to this, it binds to one site which is present within the structural genes and another site which is located very close to the lac I gene, that is the repressor gene. When this happens, the entire DNA molecule bends. This is another reason why the RNA polymerase cannot move forward and transcribe the structural

genes.

How is the Lac operon switched on if by default the Lac operon

is always switched off? How will it be switched on? It is

switched on by the effect of the allolactose. What is allolactose

? Allolactose is an intermediate which is formed when the enzyme

beta galactosidase acts on the molecule lactose. It acts on the

molecule lactose and converts it to an intermediate called allolactose. Allolactose can bind to the repressor protein.

When this binding occurs due to the binding of the allolactose, there is a confirmation or structural change which is

occurring in the repressor protein and as a result the

repressor protein can no longer bind to the operator molecule.

So when ALLOLACTOSE binds to the repressor protein the repressor

protein changes its confirmation and it is released from the

operator site. As a result the operator site is now vacant. And

RNA polymerase can proceed to.

Transcribe the structural genes,that is the ZY and the A genes.

This is one form of regulation of the lactose operon, which is

called as the negative regulation because we're dealing

with the regulator, which is repressor. Now we go to the

second type of regulation of the lactose operon, which is called as the catabolite repression. If You look carefully at the diagram over here, depicting thegenes, you will be able to see that upstream of the promoter moiety. There is another site which is present which is called as the cap binding site.

What is this cap binding site ?cap binding site is a DNA domain to which the CAP molecule binds. Cap is catabolite, activator molecule or catabolite activator protein. Catabolite activator. Protein has to bind to a cyclic AMP complex and when this complex is formed, this cap camp complex goes and binds to the cap site. When this binds to the cap site, it enhances the affinity of the RNA polymerase to the promoter region and as a result RNA polymerase binds very firmly to the promoter region and carries out transcription in a better manner or optimally.

It is not that the RNA polymerase cannot bind to the promoter in absence of Cap camp complex, it can bind. However, the transcription occurs at suboptimal levels and as a result the structural genes are not transcribed optimally and therefore the concentration of enzymes which is formed is very less. Degradation would also be very slow and very limited. Therefore it is very essential that the cap camp complex should be bound to the. Cap promoter region in order that the RNA polymerase binds optimally and carries out transcription optimally and therefore the lac or lactose operon will function at a higher level.

What is the relation of glucose to cyclic amp? The relation of glucose to cyclic amp is inversely proportional? When glucose is present in high concentrations, lactose will not be utilized by the cell. The cell will continue to utilize glucose as long as it isavailable, and only when all the glucose is depleted, then it will go on to utilization of lactose. If glucose is present in high concentrations, then it will be taken up by the cell,but it cannot be taken up as glucose. It is taken up as glucose 6 phosphate. This phosphate moiety is donated by the PTS system. That is, the Phosphoryl pyruvate phospho transferase system. In the process, the PTS gets dephosphorylated,so if more amount of glucose is taken up by the cell, then more molecules of PTS will be dephosphorylated. How does this affect the functioning of the lactose operon?

This particular phosphorylated PTS is very

important for activation of adenylate cyclase.. Adenylate cyclase

is an enzyme which helps to convert ATP to cyclic AMP, so

this conversion is done optimally only if a high

concentration of phosphorylated PTS is present within the cell.

But if the concentration of phosphorylated PTS decreases,

which happens because it donates its phosphate to glucose.

Adenylate cyclase cannot be activated an

as a result. Cyclic AMP levels also decrease, so when a lot of

glucose molecules are transported into the cell,

phosphorylated PTS levels reduce..Phosphorylated PTS cannot

activate Adenylate cyclase and cyclic AMP levels also reduce

non phosphorylated PTS. Also binds to permease enzyme

allosterically and inhibits it.Therefore, if there is a lot of

amount of glucose present then the cell will primarily utilize.

Glucose, and it will not utilize lactose and therefore the lac
Operon will be switched.

Off, but what do we mean when we say that lactose operon is

switched off? Is it like switching off a light bulb? Where either there is light or there is darkness. No, this is not so

because as we have seen earlier,we still require allolactose in

order to switch on the operon.Therefore, although we say that the lac operon has switched off, it is still functioning at

a suboptimal level. An few molecules of lactose will still

be taken up by the cell.

Which in turn can be converted to Allolactose, which in turn

can help to switch on the operon. So therefore sometimes

this is also called as a leak yoperon, or this is also called

as a leaky mechanism.

So today we have learned about the structure of the lactose

operon and the regulation of the lactose operon at 2 levels. A

negative regulation with the help of the repressor protein,

And a positive regulation with the help of the Cap camp complex

that is catabolite activator protein cyclic AMP

complex. Thank you.