Molecular Biology
Prof. Vishal Trivedi
Department of Biosciences and Bioengineering
Indian Institute of Technology, Guwahati
Module - 04
Central Dogma of Molecular Biology
Lecture-18 Genetic Material (Part 2)

Hello, everyone. This is Dr. Vishal Trivedi from Department of Biosciences and Bioengineering, IIT Guwahati and what we were discussing, we were discussing about the different properties in the course molecular biology. So far what we have discussed, we have discussed about the basic properties of the cells and then we have in the previous module we were discussing about the different types of biomolecules and we have discussed about DNA, we have discussed about the proteins, we have discussed about the enzymes and in this content in the current module we were discussing about the genomic DNA or the genomic genetic material actually. So, if you recall in the previous lecture we have discussed about the different types of experiments, how the people have figured out that which biomolecule has the potential to carry the information from the one generation to the next generation and continuing this discussion we are now going to discuss more about the genetic material and what is the makeup of genetic material and how the genetic material is actually going to be packed within the cell. So, in today's lecture we are going to discuss about the genetic material and how it is actually going to be packed into the tiny structure which is called as nucleus in the case of eukaryotic cell and in the case of prokaryotic cell it is going to be packed into a non-nucleus structure also.

So, when we talk about the genome or when we talk about the genetic material. So, the first question comes what is the genetic material? So, genetic material is a complete set of DNA comprising of nuclear and mitochondrial DNA in an organism and that is mean collectively mean called as the genetic material. This is definitely not the acceptable definitions as far as the prokaryotic is concerned because the prokaryotic cells does not contain the mitochondria. So, we are actually going to discuss about the prokaryotic structures, but in general the most acceptable definition about the genome is that it is a complete set of DNA comprising of the nuclear and as well as the mitochondrial DNA which means it is actually the DNA or it is actually the complete set of DNA which is going to be present inside the particular type of cell irrespective of whether it is a prokaryotic cell or a eukaryotic cell.

It is hereditary material which is present in an organism. So, the main purpose of the genome is that it is actually going to carry the information from the one generation to the next generation. If you recall in our previous lecture we discussed about the different

types of traits and so on although this particular course is not allowing us to discuss about the Mendel's experiments and we are not discussing about the genetic information how it is flowing from one generation to the next generation what are the different laws which are governing those kind of you know the movement of the genetic material from one generation to the next generation and so on. But it is actually the hereditary material what is present inside an organism irrespective of whether it is a prokaryotic cell or the eukaryotic cell. In the previous lecture if you recall we said that right it could be a DNA or it could be RNA.

So, because in the case of so many organisms it is could be a DNA or it could be RNA. So, the genome is totality of chromosome unique to a particular organism or any cell within the organisms. Each genome contains all of the information needed to build and maintain that particular organism. So, this is also very very important points actually there are two important point here one is that it is actually hereditary material what is present inside an organism and the second is that it is actually going to contain all the information needed for an organism to build and maintain that organism which means it is actually going to have all the informations of even about the developmental stages also how the person is how the organism will go through with the different developmental stages it is actually going to have those kind of information also. So that the organism is actually going to have the required changes in the body and so on.

For example, in the humans right in humans you are actually going to born as a baby and then you are going to have the different stages of the developmental stages and then you will reach to the you know reach to the puberty and then post puberty you are going to be adult. So, and even before birth also there are so many developmental stages you are actually going to go through and all these developmental stages are completely been governed by the genome what is present inside the organisms. Now the first question comes is how the genome and the genotype is differs right. So, there are many times the student get confused whether what is the genome and what is genotype right. So, genome is actually the hereditary material or the total hereditary material what is present in an organism is called as genome whereas, a part of the genome is actually been called as genotype.

The information contained within the chromosome or I will say a part of the chromosome this actually been called as the genotype. For example, you can have the genotype for tallness you can have the genotype for dark skins you can have the genotype for other brown eyes you can have the genotype for grey hairs and so on. So, these are the some of the properties which are actually going to be localized within a small portion of the genome they are not going to be completely there will be a genome for that particular thing right. So, genome is a collection of genotypes and the genotype

is a subset of that particular genome. Now, the question comes what are the different types of genome what are present in the different organisms.

So, we have the four categories of the genome which is according to the organisms you can have the prokaryotic organisms such as the bacteria you can have the eukaryotic organisms such as the animal and this is plant and then you can also have the specialized type of organisms such as the virus ok. So, in the case of bacteria or in general though prokaryotic organisms you can have the prokaryotic genome which is actually going to be the double standard DNA circular chromosome and then it also going to have the nucleoid. All these we are going to discuss in detail whereas, in the case of eukaryotic genome which is present in the animal you are going to have the double standard DNA you are going to have the linear DNA and it actually going to be present in the form of many chromosomes and all these are actually going to be present inside a confined structure which is called as the nucleus. So, this is the nucleus what is present right and then in the case of plants you are going to have the main genome what is present inside the nucleus just like as the animal cell and then you are also going to have the organellal genome which is going to be present in the mitochondria and the chloroplast. And then we have the viruses so, we can have the viral genome.

So, viral genome could be single standard DNA double standard DNA or it could be RNA it could be circular or it could be linear then it is segmented or non-segmented and then it is actually going to be monopartite or the multipartite. So, all these are the summary of the properties of these genome and the genome are actually been organized inside a particular organism right. For example, in the prokaryotic cell the genome is going to be distributed or going to be present within the cytosol whereas, in the case of eukaryotic cell the genome is either going to be present inside the organelles or it is actually going to be present inside a well defined structure which is called as nucleus. So, how the genome is organized? So, genome genetic organization is like that. So, in the cell the each DNA molecule associated with the protein molecules and the each DNA molecule and its associated protein is called as the chromosome.

So, in any organisms the DNA is actually going to be get associated with the protein molecules and this particular structure is going to be called as chromosomes. This organization is actually going to be hold true for prokaryotic cell or the eukaryotic cell. In the eukaryotic cell you are going to have the many types of chromosomes and the prokaryotic cell the genome is very small. So, it is actually going to have the single chromosome. So, this is just a classical example right you are going to have the eukaryotic cell you are going to have the nucleus within the nucleus you are going to have the DNA right.

So, this is the DNA right and then this DNA is actually going to be associated with the different types of proteins and that is how the DNA is actually going to be condensed in the form of different condensation organizations levels and then ultimately it is actually going to form the chromosomes. This is the similar kind of organization even in the prokaryotes the only difference is that the prokaryotes will have only one chromosome which is a circular chromosomes. And the packaging of DNA into the chromosomes. So, the DNA is actually going to be packed into a dense material and that is going to be called as chromosome. The chromosome is actually going to have the DNA and it also going to have the protein and protein is actually playing a very crucial role in packaging the DNA into the form of chromosomes.

So, the packaging of DNA into the chromosomes serves several important features. Chromosome is compact form of DNA that readily fits into the cell right this is anyway we are going to discuss in detail. Then it protects the DNA from the damages and it only packed DNA can be transmitted efficiently to go to the daughter cell when a cell divides. So, since it is a packet you can actually be able to share these packets between the daughter cells very precisely. If it is a loose DNA then it could be possibility there is a possibility that you may actually share the 50 percent, 75 percent, 80 percent like that.

But if it is a packet you will either share the complete packet or you will not share the packet. So, that actually gives a regular flexibility as well as in the regulation that the DNA is actually going to pack into the form of a chromosome. Now the question comes why there is a need to pack the DNA. So, why the packaging of DNA is required? So, DNA is packaged into a form of chromosome and then these packaged DNA is actually going to be required for many reasons. Number one it is actually going to be required for DNA competition, it is actually been required for the DNA protections, it is actually been required for regulation of the gene expression, it is also required for facilitating the DNA replication and repair.

Then also ensure the accurate chromosomal segregations and then lastly it is also required for enabling the regulatory interactions. So, what is mean by the DNA compactation? So, DNA competition by packaging the DNA into the compact structures such as the nucleosome and the higher order chromatin fibers, the physical size of the DNA molecule is reduced significantly. You know that for example, the human genome right, human genome is approximately been of a size of 1 meter fiber right. So, if you have a 1 meter fiber and you know the size of the cell right, cell is approximately 30 micrometer right. So, if you have a cell of 30 micrometer and if you have a genome of 1 meter fiber, it cannot fit into this right, it cannot be fit into this.

So, to fit this you are actually required to compact this to pack it such a dense material

that it should actually fit into this particular site. So, that is the purpose of packaging the DNA into the chromosomes. Then the second is that it is actually going to provide the protection to the DNA. So, the number 2 is it is actually going to provide the protection into the DNA. So, the densely packed chromatin structure shielded the DNA from the exposure to the potentially harmful agents such as chemicals, radiations and enzyme.

It also help to prevent the DNA from becoming tangled or breaking during the cellular processes. So, you can imagine that if I have a DNA which is loose DNA right, if it is a DNA which is loose it is actually been accessible for all sort of damaging material. For example, if you are taking a you know if you are getting exposed to the free radicals or if you are getting exposed to the hydrogen peroxide, it is actually going to have the direct access to the DNA. And this anyway we are going to discuss in our subsequent module when we are going to discuss about the DNA damage and repair. But so, there are several type of DNA damaging agents right, one is the free radicals, the other could be alkylating agents, the drugs what you are taking and all that.

So, if you are taking a drug and if the DNA is not properly packed, it will getting exposed and that is how it is actually going to be get damaged right. It is going to be damaged because there are you know modifications in the nucleotide and so on. This is completely been protected by if you have the DNA and if this DNA is been surrounded by the protein molecules. So, now what you have is if you have even these molecules, they will actually going to interact with the protein rather than the DNA. So, the drug will go and interact with the DNA because if the DNA is not going to be accessible because of it is surrounded by the different types of proteins and that is how the DNA is getting protected.

Now, the third point is the regulation of the gene expressions right. You know that the gene expression is a very tightly regulated process right. So, if the gene regulation is not been done, then it is actually going to have a very very significant negative effects on the health of that particular cell. For example, you have taken the food right and you have taken the meal right, you have taken the food, it has produced the glucose right or and the glucose the blood it has increased the blood glucose level. Now, if I have to tackle this problem, what I have to do is I have to secrete and I have to synthesize a large quantity of insulin right.

That means, as soon as this occurs, I have to do a gene expression profiling, I have to change the gene expression profiling within the pancreas and within the pancreas and as a result what will happen is the pancreatic beta cell are actually going to start secreting the insulin. That is actually going to affect on to the some of the effector organs like such as liver and muscles and that is how they will actually going to convert the glucose into

the glycogen right and that is how they are actually going to protect the body from the harmful effects of having the very high level of blood glucose. Now, this is temporary right, this effect is temporary because after some time the blood glucose level will reach to a normal level and then if this process will continue, then it will actually going to go down to the liver right. For example, if the blood glucose level is 80 milligrams per deciliter right, which means 80 milligrams per 100 ml, then it is the normal level right. But when as soon as you have taken the food, the level will go down go up right, level will go up to like for example 200 right and from 200 it will return back to 80.

But if this process will continue, it will further come down right, it will come down to 50 right and it will come down to 0 if this will continue because the insulin does not know that there is a glucose, there is enough amount of glucose right. So then so there is a regulation of gene expression required right, as soon as this reaches to 80, then the blood glucose level or it is actually going to give the indication to the pancreatic cell that ok, there is no more insulin required and that is how it is actually going to change again, it is actually going to change the gene expression profiling and that is how there will be no secretion of insulin. And that is why it is very important for maintaining the normal physiology of an organisms. So regulation of gene expression, the DNA packaging in can influence the accessibility of the gene to the cellular machinery involved in the gene expression, such as the transcription factor and RNA polymerase. By compacting or loosening the chromatin structure, cells can control which genes are accessible for transcription and thus regulate the gene expression pattern.

So we have discussed how the gene expression profiling is going to have the significant effect on to the overall physiology of that particular organism and it is been influenced completely by whether the DNA is present in a compact structure or not. Because if it is if the gene is present in a compact structure and it is not accessible for the cellular machinery to perform the transcription and translation, then that will not going to be transcribed. On the other hand, as soon as you would like to have the down regulation of a particular gene expression, you just put that particular gene into a tight compact structure and that is how it is actually going to control the overall gene expression of that particular protein. Now, the fourth point is it is actually going to facilitate the DNA replication and repair. This anyway we are going to discuss in detail when we are going to talk about the DNA damage and repair and all that and replication also.

So during the DNA replication and repair process, the packaging of DNA into the nucleosome must be temporarily loosened to allow the necessary protein and enzyme to access the DNA strands. After replication or repair, DNA is repacked into the nucleosome and hired or chromatin structures. Hence, the proper genome packaging ensure accurate replication and repair of DNA. So this is very very important that we

should have the packaging and the unpackaging of the chromosomes so that the some amount of DNA is going to be open and then that DNA is going to be replicated.

Now the number of 0.5 is it is actually going to ensure the accurate chromatin segregations. So during the cell division, the genome must be accurately divided between the daughter cells, the compact organization of the DNA into the chromosome facilitate this process. This anyway we have discussed in detail that if you have a single chromosome, you will divide this chromosome and you will make two chromosomes. And then you will actually going to divide this chromosomes equally. You are going to take give one to the sister or the daughter cell and one you are going to keep it along with the parents.

If it is not compact, if it is a DNA, if it is a DNA, if it is not compact, if it is a DNA, then the division could be 70, 80 or percent. It could vary because some amount of DNA you will put it into a sister or daughter cell, some amount of DNA you will put it into parent cell and so on. So there are possibility that the daughter cells will have 1.25 copies of the genome and the parents will only left with the 75 percent. So this kind of possibility should not exist that is why the DNA is actually going to be packed into a packets.

So you will just take one packet with you among yourself and then you are actually going to give one packet to the daughter cell and that is how there will be an equal division of the genetic material between the two daughter cells. Then we have the enables the regulatory interactions which is the point next point. The 3D organization of the genome within the nucleus allow for the specific regulatory interaction between the different region of DNA. This is special organization facilitate the long range interaction between the enhancer region of DNA. So this is all about why the DNA is required to be packed into a compact structure to form the chromosomes.

Now let us see how you can be having this kind of organized this kind of packaging into the different types of organisms. So we will start with the prokaryotic organisms. So first we are going to discuss how the genome is going to be organized into the prokaryotic structures. So the genome organization in a prokaryotic cell. So they these are this is the typical prokaryotic cell where you have the cell wall you are going to have the capsule you are going to have the plasma membrane and inside the plasma membrane you are going to have the cytosol and then you are going to have the nucleoid.

So nucleoid is the region where you the all the genomic content is going to be present and then you also have the ribosomes and the plasmids and you are going to have the all this kind of phagella and all that. So they have these they have these small bodies and a

small genome. You know that the genome bacterial is bacteria cells are very small. So they are going to have the smaller genomes. They do not contain a nucleus or any other membrane bound organ.

This all we have discussed when we were talking about the cellular structures. Then they have a small circular DNA which is present inside the nucleoid region. So this is the nucleoid region where the genomic content of the prokaryotic cell is going to be present. They have a single chromosome that floats within the cytoplasm. The genome size ranges between 10 to power 4 to 10 to power 7 base pair with a high gene density.

Apart from this single chromosome some bacteria have the extra chromosomal DNA which is called as the plasmids. We have also discussed about the plasmids when we were talking about the bacterial cell and we have also discussed how you can be able to isolate the plasmids from the bacterial cell and we have shown you a demo video also how to do that. So in a prokaryotic cell you going to have the chromosomal DNA as the genomic content. The second is you are going to have the extra chromosomal DNA which is also been called as plasmids.

So let us discuss about the plasmids. So prokaryotic frequently carry one or more smaller independent extra chromosomal DNA which is called as plasmids. Plasmids are not genomic DNA they are accessory DNA molecules. A smaller circular DNA molecule that have the ability to self replicate. Unlike the larger chromosomal DNA plasmids typically are not essential for the bacterial growth. So plasmids are actually been required for providing the specific properties and by the plasmids they can be able to exchange that property between the particular bacterial colony.

For example there could be a property like resistance against ampicillin. So if a bacterial colony has acquired a resistance against the ampicillin and that property it has captured in the form of a plasmid then it actually can share that plasmid within the colony and that is how they can be able to distribute that particular property within the organisms. What is the importance of the plasmids? So plasmids provide advantage to the bacteria such as antibiotic resistance, herbicide resistance etc. So all these properties are actually been due to the different types of antibiotic, different types of genes and all these genes are actually going to be cloned within the plasmids and that is how the plasmids are actually going to express the protein and provide the necessarily resistance mechanisms within the cell. And that is how the bacteria can actually provide that particular plasmid to the colony and that is how the colony is actually going to be resistance.

In addition unlike chromosomal DNA plasmids are often present in many complete

copies per cell. So unlike the chromosomal DNA the plasmid will not have the bacteria will not have the single colony single copy of the plasmid it could have the 200 copies it could have 500 copies and so on because the number of copies will decide which bacteria will have the higher resistance property. So if you have a very high number of plasmids you can be able to have the higher resistance for that particular antibiotic or that particular type of phenotype. Then we will talk about the bacterial genome.

So bacterial genome is very small. So bacterial chromosomal DNA is usually a circular molecule that has a few million nucleotides in length. For example in the case of E. coli you have the 4.6 million base pairs. Similarly you have the H influenza H influenza is going to have 1.

8 million copies. So it is actually a small genome what is present in the bacteria. And then a typical bacterial chromosome contain a few thousands different types of genes. So the unlike the eukaryotic organisms you are not going to have the useless genes you are not going to have the other kinds of non intron or non expression genes actually. So bacteria only contains that gene which are going to be expressed and which are actually going to have some meaningful effect or meaningful purpose inside the cell because you know that they have their size is very small so they do not want to cover or they do not want to keep the unwanted materials. Then structural gene sequence account for the majority of the bacterial DNA or the encoding proteins.

The untranscribed DNA between the adjacent genes are termed as intergenic regions and these process these regions are very very small or almost absent in the bacterial system. Then the since you have the DNA you have to pack this DNA into form of a chromosome so that you can be able to make the compactation and make make the structure very compact. So packaging of DNA so prokaryotic cell usually have a smaller genome that need to pack their DNA is still substantial you know that we bacteria are few mu micron in size so they are DNA size is also relatively very big so it has to be compacted. E. coli must pack its 1 mm chromosome into a cell that is only 1 micrometer in length.

It is less clear how the prokaryotic DNA compacted but it is actually going to be packed into a small structure or within the cell. So the region what it is actually going to pack the DNA is called as nucleoid. So nucleoid is a primitive nucleus or I will say it is actually a primitive nucleus except that it is not going to have the membrane. So it is not a membrane it is a region in which the bacteria is actually going to have the chromosomal DNA. A prokaryotic chromosome is circular and it is reside in a cell region called as the nucleoid.

Only one complete copy of their chromosome that is packed into a nucleoid. 80% DNA by maze can be unfolded by agent that act on RNA or the protein. The proteins responsible for condensation and maintaining the super coiled structure of the DNA have not been identified. So that is still unknown right how the different types of proteins are involved and what are the different types of proteins are involved into the making the structure very compact. The type of protein found in prokaryotic chromosome known as the nucleoid associated protein which is responsible for compacting the DNA into a chromosomal structures.

DNA determine the protein what protein and enzyme an organism can synthesize and therefore what chemical reaction it can be able to carry out. So what is the function of the genome whether in any organism that it is actually going to determine the proteome of that particular organisms. It is actually going to decide what are different types of proteins and enzyme are going to be produced and that is how it is actually going to eventually we decide the metabolism and physiology of that particular organism. So this is the micrograph of a bacterial cell where the nucleoid is actually going to be shown. So this is actually the region where the nucleoid is present and within the nucleoid what you are going to have is the bacterial chromosome which is super coiled actually.

What are the key features of the nucleoid is that most but not all bacterial species contain the circular chromosomal DNA. A typical chromosome is a few million base pair in length. Most bacterial species contain single type of chromosome but it may be present in multiple copies. Several thousand different genes are inter dispersed throughout the chromosome and one origin of replication is required to initiate the DNA replications. So this anyway we are going to discuss when you are going to discuss about the origin of applications.

So origin of applications in the case of the bacterial chromosome is single. So it is actually going to start here and it is actually going to go through and then it will come to here right and you know that it is actually going to go in both the direction. So this is going to be leading strand and this is going to be lagging strand and that is how it is actually going to produce two copies of the genome after one cycle right. One will which will come from this side and the second which will come from this side.

So second will come like this right. So that is why it is actually going to have one original copy and the one is replicated copy. The short repetitive sequence may be inter dispersed throughout the chromosomes. The chromosome DNA must be compacted about a thousand fold remember that 1 mm will actually need to be compacted within the 1 micrometer diameter. So it is actually has to be compacted around thousand folds. The formation of the loop domains so and the number of loop varies according to the size of

the bacterial chromosome and the species.

For example the E. coli has 50 to 100 with 4000 to 40,000 to 80,000 base pair of DNA in each. So you are going to have a circular chromosome and then this circular chromosome is actually going to be looped and it is actually going to compacted by doing this and that is how it is actually going to form the very strong structures. Well let us take an example of one bacterial chromosome where one bacterial species how the chromosome is occurring.

So in the case of E. coli the E. coli chromosome is compact one fifth of a volume right. The determinants of the nucleoid folding so negative super coiling by the topoisomerizes and the condensation by the attachment of the nucleotide structure proteins. The nucleoid is highly condensed during the rapid growth and RNA polymerase concentrate in the transcriptional loci and the RNA polymerase is distributed throughout the chromosome. So this is the bacterial cell where we have already discussed about the different types of parameters. So this is all about the genomic material and what we have discussed we have discussed about the importance of the genetic material and what are the different types of genetic material are present in the different types of organisms.

So we have taken an example of the perocratic structures then we have also discussed about the Eukaryotic structures and when we also discuss about the viruses. So in the case of prokaryotic you have the double standard or double standard DNA and it is going to be present in the cytosol in the form of circular chromosome and then apart from that is also going to have the extra chromosomal DNA in the form of plasmids. In the case of Eukaryotic cell you are going to have the either the genetic material present inside the nucleus or it is actually going to have the genetic material which is present inside the organelle such as the mitochondria or the chloroplast. In the case of viruses it can have the single standard DNA or the double standard DNA or the RNA and the viruses are unique organisms so they are actually going to have the different types of physiology and different types of manipulation of their genetic material. So and then lastly we have also discussed about the how the genome is organized within the prokaryotic structures and how the compactation is occurring inside the genomic inside the prokaryotic cell and so on.

So with this I would like to conclude my lecture here in our subsequent lecture we are going to discuss some more aspects related to the genomic material or the genomic content. Thank you.