Applied Environmental Microbiology
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Lecture - 05
Central Dogma I

Hello students. Welcome once again to our course on Applied Environmental Microbiology. Today, I am going to talk about what is referred to as the Central Dogma of microbiology. So, up till now we have talked about the cell structure, how it is made what are it is components and had a brief glance at their functions and properties. And I ended the previous lecture talking about genes introducing you to genetic elements and even hinting about the idea of chromosome and extra chromosomal genetic material. Today we are going to dive deeper into the informational molecule such as genes DNA nucleic acids in the cell and notice how they regulate life and this involves 3 steps; first is replication, second is transcription and third is translation and this together are known as central dogma of molecular biology.

So, today we are going to learn about first gene genetic elements and genome this is from the last lecture.

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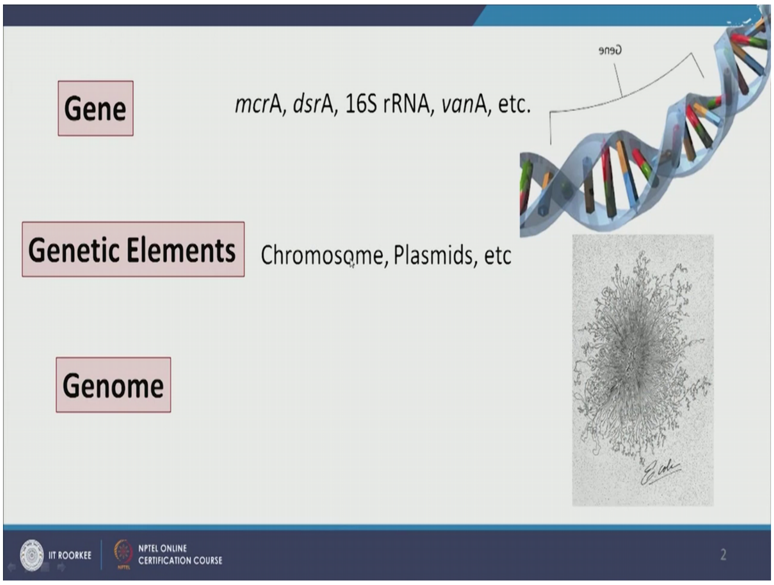

So, just as a revision gene are those fragments of nucleic acids that together make sense. The together code for a command genetic element are all the genes and non-genetic non gene genetic material together that are clustered together such as chromosomal plasmid and genome is all genetic material in a cell. So, let me illustrate this using a graph.

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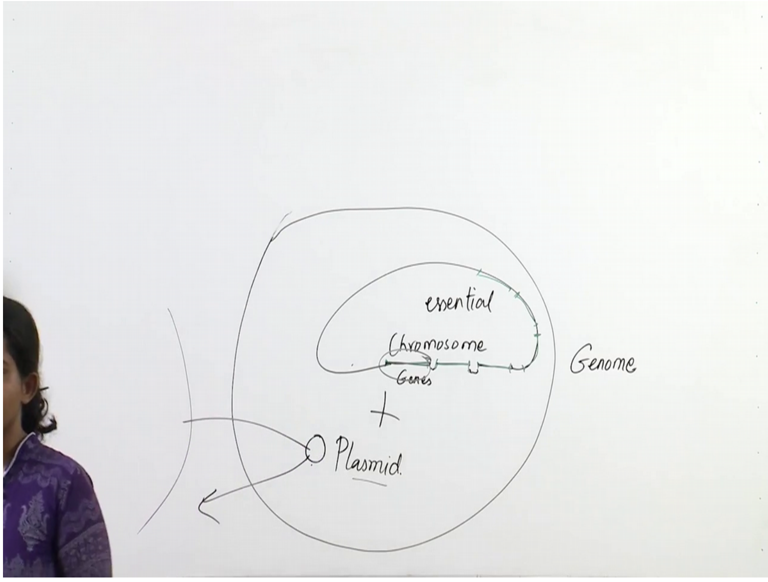

So, let us imagine this is a prokaryotic cell, they are usually easier to study because their genetic material just lies nearly suspended in their cytoplasm. So, let us saw from the very basic and we have genes.

So, each gene encodes a protein now a protein is an enzyme or a material that will allow cell to do some microbial activity. So, each of this gene in very literary terms right now we can express it as it is a sentence it is a command. Now these genes are woven together by this inter genetic material into long chains of genetic material refer to as chromosome. So, basically what we have is a chromosome is a sequence of genes separated by nucleic acids that do not necessarily code for a particular protein and this genetic material of the cell usually contains all the essential information that a cell requires to grow and to live it is life sometimes and very often in case of prokaryotes.

Microbes also have extra genetic material extra chromosomal genetic material let us say this is a plasmid is a classical example of extra chromosomal genetic material now this chromosome remember is essential. So, this is essential genetic material for the cell to grow plasmid might also carry some essential genetic material, but the beauty of plasmid is that since it is not part of chromosome it is easy for the cell to reject the plasmid throw it out of this cell or share it with another microbial cell.

So, to recap we have genes each gene encodes for a particular protein or part of a protein usually a protein, these are genetic elements plasmid chromosomes these are genetic elements together chromosome plasmid and all of the genetic material in the cell.

Together they are referred to as genome. So, when we talk about human genome we are not only talking about what is there in a chromosome, but we are also talking about the extra chromosomal genetic material present in human cells. So, now, let us go down and we will go down to the basic of what genes are what DNA is what these nucleic acids are so that it is more clear to you.

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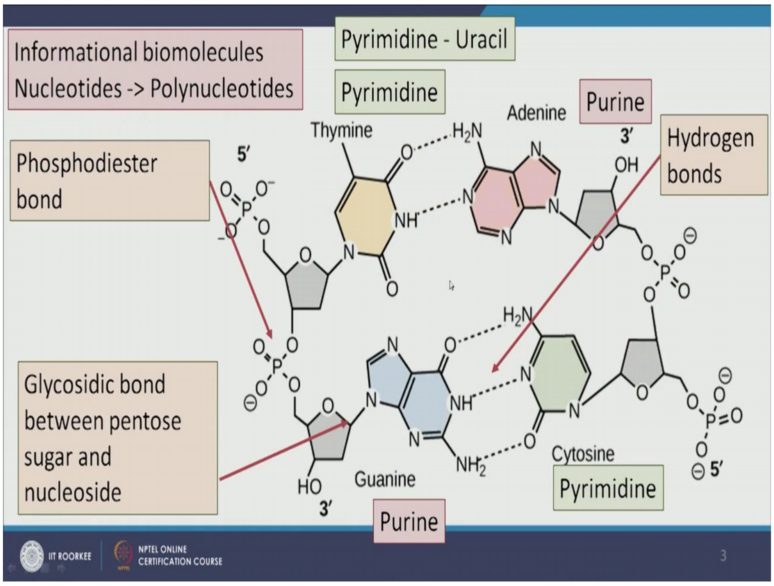

So, let us go down to the basic and look at what constitutes the genetic material. In this slide we have 4 informational molecules attached to phosphate bond phosphate backbone, cytosine Guanine thymine and adenine, thymine adenine Guanine and cytosine often abbreviated as ATGC form the basic alphabet of life.

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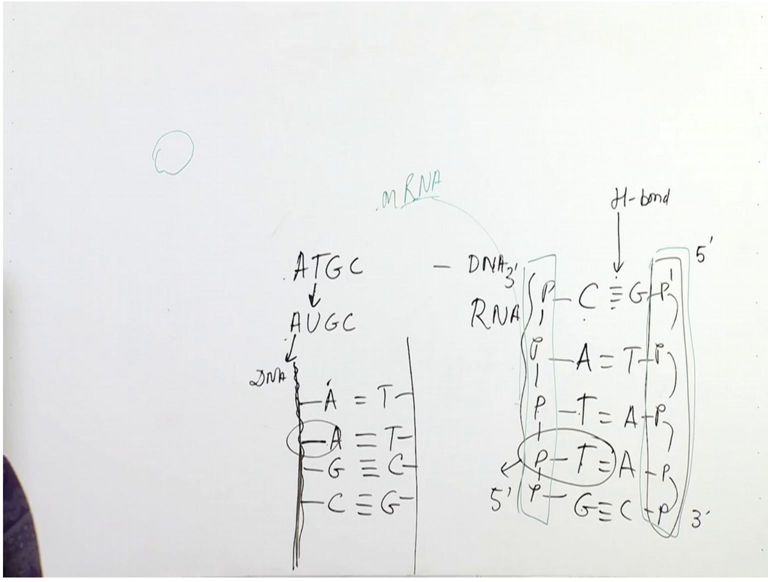

ATGC Adenine thymine Guanine cytosine these are found in DNA because they are deoxyribonucleic acids that is it chemistry by the way and when we have RNA the T is replaced by U. So, we have AUGC. So, for example, I ask you a question and I give the question is that I give you 2 sequences and I ask you to determine which of these is DNA which of these is RNA; what you need to do is look if it has T or u if it has u it is RNA. If it has T it is DNA alright. So, ATGC and AUGC now what do these ATGC and AUGC do usually these are nuclear sites and then they get attached to a phosphate backbone of DNA. Now here is a beauty a will form bonds with T these are weak hydrogen bonds.

So, if there is this is let us say our genetic material DNA because we are talking about t. So, this is our DNA 1 chain of 1 phosphate bond of DNA backbone of DNA and there is an a attached to it will automatically try to attract A T to it and let us say here is another A. So, we will have another T, here we have A G now G makes 3 hydrogen bonds with C and let us say we have C here. So, it will attract A G now these also are attached to A phosphate backbone and together they make a DNA molecule. So, this together they are DNA molecule these are nucleosides and when they are attached to the phosphate backbone they are called as nucleotides.

So, in this figure you cannot be see here that we have thymine is attached to the phosphate sugar backbone and by and they are attached to another guanine molecule that is also attached to a backbone of nucleic acid. These 2 are attached to each other with a phosphordiester bond.

Now because there is a thymine here it has a double bond with Adenine and the guanine has a triple hydrogen bond with cytosine G with C A with t and now these nucleotides the Adenine and the cytosine are attached to each other by a phosphodiester bond.

Now among ATGC or adenine thymine guanine and cytosine thymine and cytosine are pyrimidine and you can notice the uniqueness between thymine and cytosine they both have single aromatic ring single aromatic ring. There is some structural difference in both of them definitely now guanine and Adenine are called purines and you can notice they have 2 aromatic rings both of them similar structure, which is still distinct from each other. So, we have Adenine one in purine thymine cytosine pyrimidine.

Now thymine is attached to the sugar and phosphate bond this is phosphate phosphodiester bond and it is important to know that the pentose sugar by the way. Now pentose sugar and nucleoside are attached by glycosidic bond and this is hydrogen bond. So, there are 3 different bonds here there you must be aware of. So, going back here and trying to illustrate this is what I want to the message that I want you to remember A A T T G C. Let us say this is the sequence of nucleoside by themselves they referred to as nucleoside when they attach to a pentose sugar and the diagram is there in the slide the pentose sugar. So, they are attach to a pentose sugar now they are called as nucleotides these pentose sugar have phosphodiester bonds with them. So, phosphodiester so they have a phosphate molecule connecting the pentose sugar.

Now together they have made one chain one half of the DNA. Now because it is A C it will make triple bond with G A will make double bond with T and so on. And they are attached to another Pentose sugar and the pentose sugar have phosphodiester bond between them. And this is called the phosphor or pentose the sugar phosphate bond backbone of DNA now you can notice that there are 2 chains going this is a these are usually very very long chains. So, very long up to million base pairs by the way this unit is called base pair; what I just call nucleotide is called base pair.

So, we count them using base pairs for example, how many base pairs do we have here 1 2 3 4 5. So, this DNA how long is it is 5 base pair long so the unit of length of DNA for

Applied environmental microbiology is base pair. If I tell you I have sequences of that are 500 million base pair long which is very long, then it means I have information about 500 million base pairs that are connected sequentially to each other alright. Now this is the central dogma of molecular biology that I promise we will be covering in this course.

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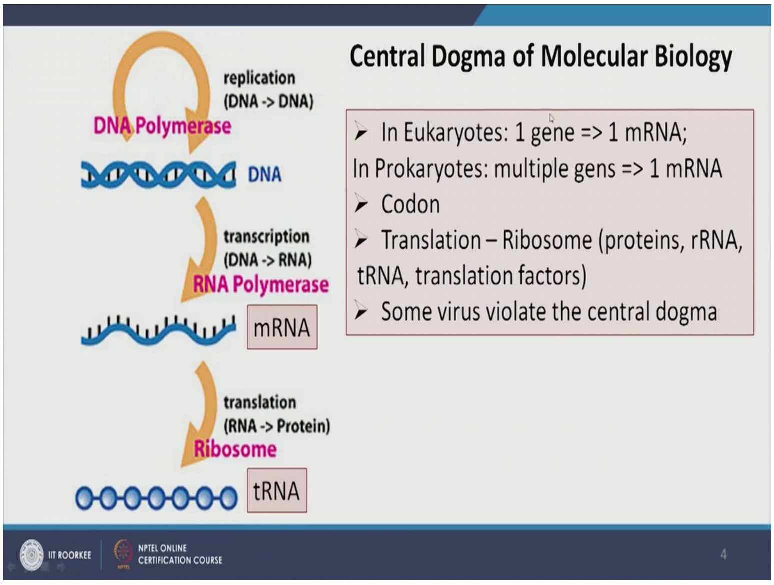

So, we have DNA now let us take some time and understand about the structure of DNA they are very generally. So, I told you that we have 2 backbones. So, this is one pentose phosphate backbone, this is another pentose phosphate backbone they do not stay to each other like parallel strips instead what they do is they twist around themselves and make what is called a double helix structure.

So, this is their helically coiled around each other and this is the beauty of it even though these are hydrogen bonds here which are very weak bonds in by their nature, because there is so many hydrogen bonds they bonding between the both backbones is very strong just because of sheer number of hydrogen bonds that are represent here. So, this is coiled like a double helix the best way to imagine a double helix is imagine a stairwell coiling up and another stairwell coiling down. So, they are anti-parallel to each other and the various anti parallel business works is for now just to make it simple we call one and 5 prime another 3 3 prime.

So, 1 stand runs 5 prime to 3 prime the other stand runs 3 prime to 5 prime. So, DNA this is DNA again reminding you. So, DNA is a double helically coiled structure with 2 phospho pentose backbone and they have nucleosides that attach to it and bond to each other um such that t is always bonded to A and G is always bonded to C by double and triple bond and they are anti-parallel in sense that one runs from 3 prime to 5 prime the other runs from 5 prime to 3 prime. And briefly glancing the central dogma of molecular biology this DNA that we just described here in detail is transcribed into RNA messenger RNA.

Now what is transcription DNA acts like the storage of information and the information has to be taken to other parts of the cell. So, there all this is happening in the genetic material now the cell let us say the entire white board is the cell. So, let us say here is a ribosome that needs a particular protein or the cell needs to make a particular protein. So, the message needs to reach the ribosome. So, the way it will work is this particular copy of DNA will be photocopied will be replicated into a messenger RNA.

Now remember what I told you in the beginning because it is RNA instead of T it will have U. So, when up cell a bacterial cell even eukaryotic cell looks and finds that this genetic material has u it knows that it is a message it is a messenger RNA. So, because it is a message let us take action on it and then the cell might decide then MRNA can go where it needs to go and then the cell might decide to act on this message .

So, this step where DNA is converted into RNA or messenger RNA is called transcription and then the mRNA reaches the ribosome where synergistic efforts of ribosome and other RNAS transfer RNA and proteins together convert this messenger RNA use this messenger RNA as a template for making a new protein.

So, remember central dogma of microbiology a molecular biology is we have DNA that encodes information from this information we can make copies of genetic material called messenger RNA this carry the information encoded in DNA and the messenger RNA can go to ribosome, which can make protein using mRNA as a template and say beautiful mechanism and I will be discussing more details about it as we now, just basic information and eukaryotes usually we have one gene let us say from here to here it is 1 gene. Now in eukaryotes this one gene will be converted into 1 mRNA 1 messenger RNA, in prokaryote let us say this is 1 gene this another gene here and there 4 more genes and this side and 2 more genes on this side now all these 8 genes can be can be can be transcribed into a singular long mRNA. So, in prokaryotes you can see transcription can happen very fast and energetically very efficiently now this mRNA goes to ribosome.

Now, as I said ribosome involves similar just a take efforts of ribosome proteins translation factors tRNA. So, here we have information saying that ribosome which in which is; may constituted of proteins and r RNA ribosomal RNA it makes use of some transfer RNA or tRNA and translation factors to convert to create proteins using this as template. This is a very beautiful mechanism and I would like to draw this and show it to you. So, now, let us take an illustrative insight into the central dogma of molecular biology. So, as I said we have DNA.

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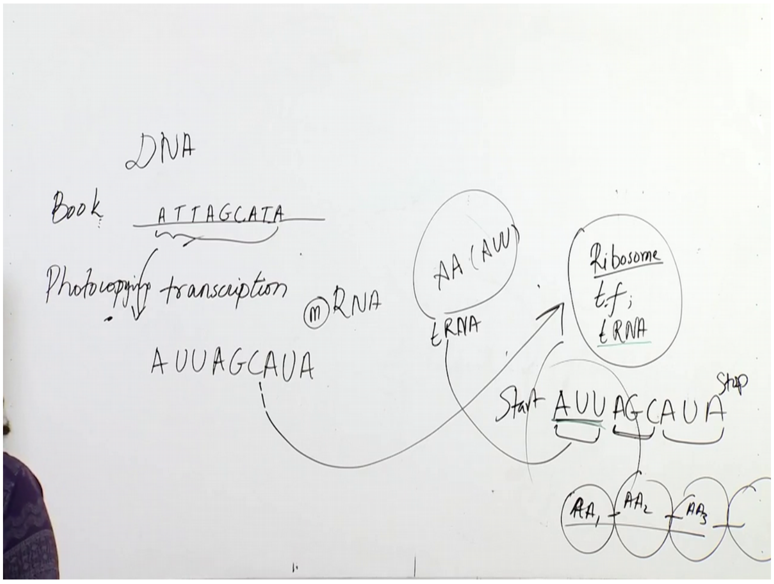

So, first we have DNA made of ATGC and a beautiful phosphate pentose backbone and double helically coiled antiviral sends to each other. Now the DNA let us say the sequence of DNA looks like this A T T A G C A T A and so on and so forth. Let us say this is in this is an extremely small gene that needs to be sent to some part of the cell. So, that action can be taken according to the information stored here in a layman's term I like to call DNA as the book in a library.

So, if there is a book in the library that the information is that is needed in some part of the campus yours university campus for example, then first step is to go and find the book. So, if let us say I found the book and I know that this is the page gene is the page that is important for you this is the page that I need to do the work that I need to do at some other part of the campus.

So, what this is what the next step that will happen is called as transcription. So, in transcription that DNA is converted into RNA messenger RNA and it is called messenger RNA, because it carries a message to the cell that hey this is the page you require and this is the page from the book that I got I like to think of transcription as photocopying the page in the book. So, let us imagine that you are sitting in your university campus and you find out that you need to make a septic tank, because it is environmental microbiology let us talk about septic tank you need to make a septic tank and you do not have the information how to make a septic tank.

So, what you can do is you can go to a library find the right book that has information. So, you find the right gene that has the information you require about how to make a septic tank then you can go to photocopier and ask him to make photocopy of the pages that you require this is transcription. So, DNA is converted into messenger RNA. So, the messenger RNA for this would look like A U U A G C A U A. So, this has been converted into messenger RNA now this messenger RNA is shorter than the DNA the entire DNA genetic material and this can be taken wherever you want it to go. So, now, I take the photocopy of the pages that I required to make a septic tank and I can go to the site where I need to make a septic tank.

So, this will go to the site now in on the side we have civil engineers, we have architects for some reason we have economists, we have masons and we have the construction project managers. So, we have a group of people now in terms of molecular biology this people are called as ribosome. So, we have ribosome which is a which is an elegant structure of different kinds of RNA proteins and if you take a generic course and genetics you get to learn more about ribosome and I highly encourage it is. So, very beautiful to learn about ribosome and it is a very very important part of cell.

So, this is where the meeting is going on the meeting of construction project managers and the architects and the civil engineers for making your septic tank in terms of molecular biology. This is where the action will start taking place this is where your; the copy that you have made from some pages of the book will be converted into action. So, we here we have ribosome we have translation factors we have a tRNA or transfer RNA and we have loads of other proteins. Now what they do is they have received a message this messenger RNA has gone here they have received a message and the let us bring message here now A W. So, they have received your messages now they will read your message. So, how does cell read the message, usually every gene will have a start codon. So, that says start; start reading message from here and it will have a stop codon that will say stop; stop reading message here. So, wherever the ribosome and their ribosome and company find a start codon there started in codon. Now here is the thing in cell reads in terms of 3 alphabets at a time.

So, it will read A U U A G C A U A what it will do now is bring the tR trans station factor and ribosome will stick here and they will read A U U. So, when they have read A U U the transfer RNA will bring the right amino acid that matches with A U U. Now I must pause here and introduce you with what this amino acid is and what it is tRNA is doing here.

There are amino acids that join together with peptide bonds and make protein some of them are essential amino acid some of them are non-essential, but some of them can be created in our bodies in ourselves and we need acquire for environment. Now these amino acids for each amino acid usually there is a unique codon and we will get there I will show you the chart there is a unique set of 3 nucleotides that say I want this amino acid. And what transfer RNA will do when ribosome has read oh the code says A U U then the transfer RNA that carries the right amino acid which is encoded by A U U will come here and will bring amino acid.

And then when the ribosome proceeds further and reads AGC another transfer RNA carrying the right amino acid that is encoded by AGC will bring that amino acid and they will join together by peptide bonds in this way they make long chains of amino acid now it is important to know that these long chains of amino acid are the action this is at the actual action that was encoded by your DNA in the first place. So, now, these proteins will do the work they need to do they might catalyse a reaction they might trigger some actions in the cell or improve the met or change the metabolism of the cell or whatever the function that is there of this protein, now it is beautiful for me to just mention I want to mention this beautiful aspect of proteins that it is not their chemistry per say.

It is not protein one amino acid 1 here and then we have amino acid 2 here and amino acid 3 here it is not the sequence of amino acids like at a sequence of mRNA or sequence of nucleotides in DNA that governs a function of protein it is their stereochemistry not just chemistry and I will talk about it soon. So, here we have central dogma of molecular biology and these are some pictures beautiful cartoons showing what I just described to you on the board.

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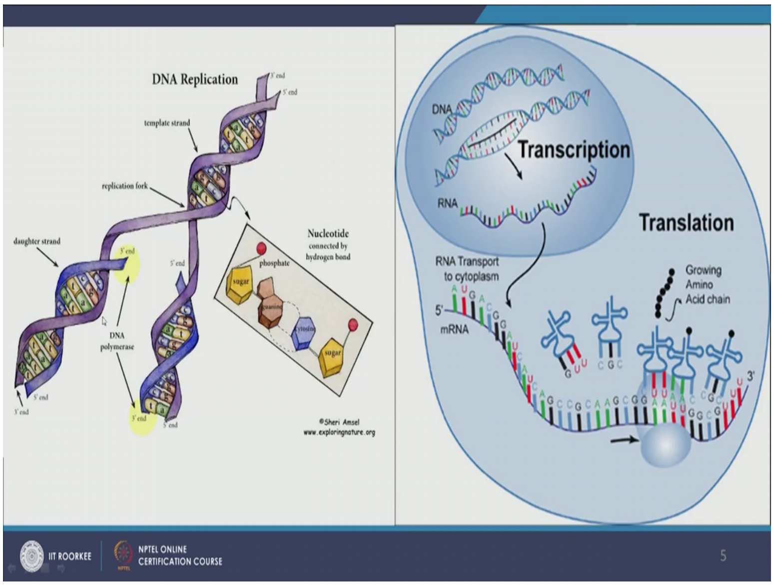

So, we have in central dogma the first step is DNA replication which is step that I skipped.

So, in DNA replication if the cell wants to divide it can use topoisomerase, which is a protein which breaks and uncoil the DNA and then when the coils of the DNA have been this have been separated new complementary nucleotides can attach and make the complementary DNA chain.

So, in this case in this purple backbone has been torn apart and when it was torn apart you can see in this 5 prime end of this purple backbone there was an A followed by G then G and followed by C. So, for an A or T came and attached to it than A C came an attached to it and then C came in attached to it and so and so forth and a complimentary chain is being formed.

And this Job is done by DNA polymerase by the way. So, this is the first step if we want to replicate a cell we do DNA replication and do not need to go more in detail, but you should know the DNA polymerase is at work it is important to know because many chemicals stop the activity of DNA polymerase and the cells die the second step is transcription. So, here we have DNA now we want to make messenger RNA now to make messenger RNA topoisomerase will come in to the uncoil this DNA. I must mention here that DNA can be very long for a cell like e coli which is very very tiny, it is DNA can be up to 1.8 millimeter long.

Now how would you pack such a long DNA in such a small cell by super coiling, which is the; this DNA is super coiled to an extended there is actually a lot of force lot of tension stored in it. So, taupe I somewhere is a very magnificent protein structure will come here uncoil it and when it opens up of required fragment of DNA as shown in this part of the figure complementary strands will come and will be transcribed and what we get is a beautiful is a single stranded RNA. If you remember in DNA we have ATGC in RNA we have AUGC. Now this messenger RNA will go to ribosome and this is the picture of ribosome here it is like blue 2 circles. Now the ribosome will read the messenger RNA and it will read it in terms of 3 codes 3 nucleotides together they are referred to as codon.

So, first it write G U U here and the transfer RNA this structure is of transfer RNA the transfer RNA that carries a mirror image of G U U which is C A A and the right amino acid. So, this black dot is the amino acid. So, this is right amino acid will come and attach to this and it will write amino acid will come and attached to the growing amino acid chain or the growing protein change this is transcription followed by translation and this amino acid is the enzyme required for the activity. So, we can see here how the information converted into action and in the analogy that I was drawing about library if this is the book in the library transcription is making a copy of a particular important page and then you take the page to where you where the action is happening and you convert that information in the page into activity which is protein so.

Now, you know 3 kinds of nucleic acids and I want to give you a quick glimpse on their properties DNA if it is double stranded. So, this is double stranded DNA right 2 strands that are super coiled and helically arranged helically attached to each other. So, this DNA is usually very long lasting unless there are specific enzymes that degrade DNA the DNA can last very long and there we have even managed to collect DNA from certain extinct animals that died a long long time ago. So, you know DNA can lost thousands and thousands of years.

RNA on the other hand usually has a short life and you can see why DNA needs to sit here forever and ever, but RNA needs to be destroyed once the protein has been made if let us say the RNA encodes for septic tank well it would not, but in human life it is it is encodes for a septic tank then unless we destroy this people will continue making more and more septic tank.

So, in terms of molecular biology RNA needs to have a particular life lifespan if it lives longer than it is lifespan, then the same protein will be unnecessarily made wasting cells resources and also perhaps causing damage to the cell on the other hand if mRNA dies really fast then the required action will not happen, now here I have a very interesting bullet point saying some virus violate the central dogma some viruses while the central dogma because they skip the DNA step entirely they just have RNA these are RNA viruses and we will briefly go over them in the later chapters.

And then the diversity among domains here I want to highlight the immense diversity in the ribosomes.

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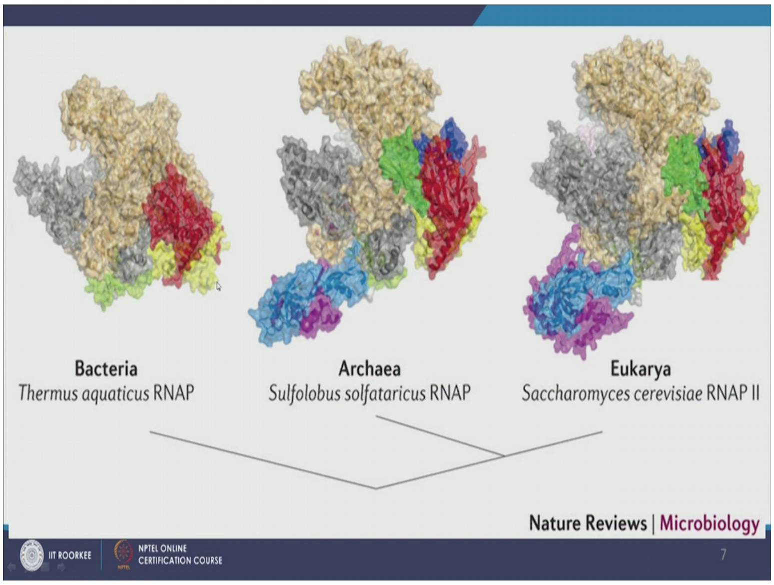

So, let us take a look here in this picture we have bacterial ribosome Archaeal ribosome and Eukaryotic ribosome and picture suggest that the eukaryotic ribosomes are most complex in structure and bacterial ribosomes are simpler and have less components. What we notice is that the ribosome activity of Archaea is usually the most simple compared to bacteria in Eukarya in eukaryotic cells this entire activity that is done by ribosome register of translation is actually have 3 different steps. So, it takes 3 unique steps for eukaryote to convert message into action message into protein Archaea usually has a simpler structure and bacterial ribosome has been very very useful for taxonomic classification which simply means giving bacteria a right name and knowing the bacteria that it is related to.

So, if I have an unidentified bacteria and I have the sequence of it is ribosome because rainbow ribosome was most of has nucleic acids RNA. So, if I have the sequence of RNA I can find out which other bacteria it is related to and not and we will talk about this in subsequent lectures.

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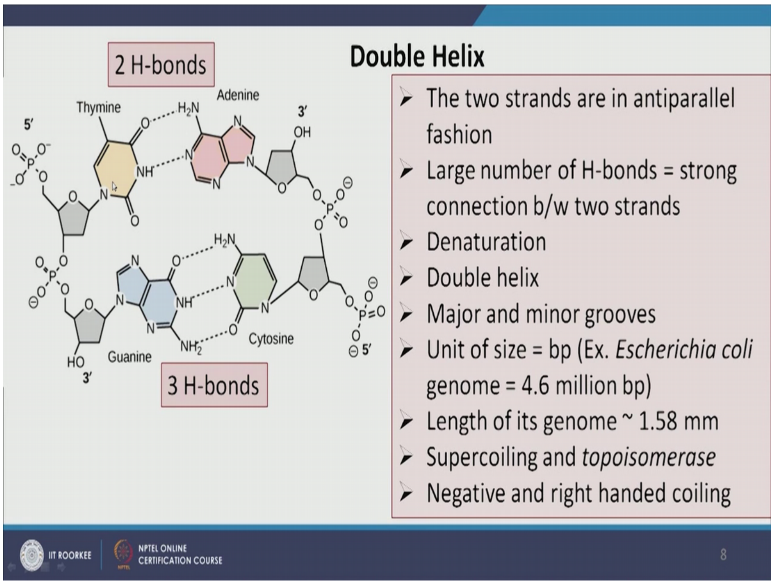

I have mentioned to you before that T and 8 bonds to each other with double hydrogen bonds and guanine and cytosine with triple hydrogen bonds. Now this bonding can be very strong not because they are hydrogen bonds which are in general very weak bonds, but because there are so many bonds hydrogen bonds between the 2 strands of DNA.

Now, if we increase the temperature now what happens when we increase the temperature students when we increase the temperature, the energy of the heat can dismantle these bonds and what we get is denaturation which is the strands separate from each other.

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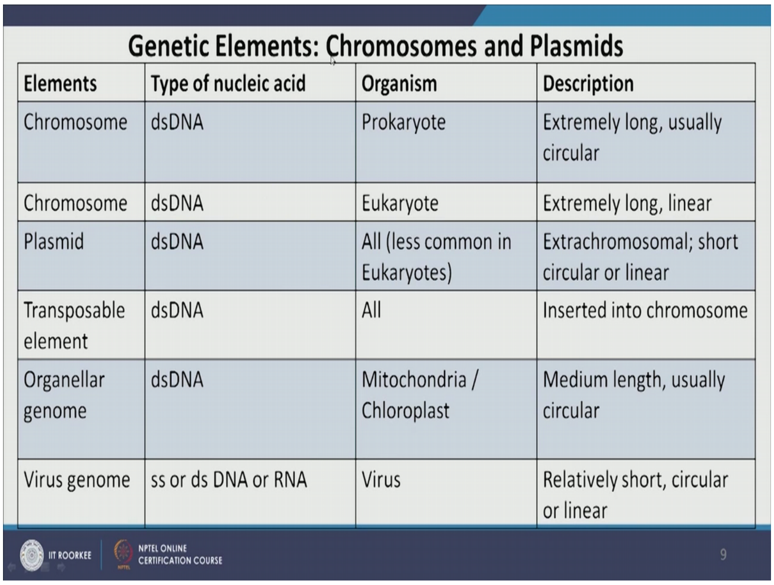

Genetic Elements: Chromosomes and Plasmids

Elements	Type of nucleic acid	Organism	Description
Chromosome	dsDNA	Prokaryote	Extremely long, usually circular
Chromosome	dsDNA	Eukaryote	Extremely long, linear
Plasmid	dsDNA	All (less common in Eukaryotes)	Extrachromosomal; short circular or linear
Transposable element	dsDNA	All	Inserted into chromosome
Organellar genome	dsDNA	Mitochondria / Chloroplast	Medium length, usually circular
Virus genome	ss or ds DNA or RNA	Virus	Relatively short, circular or linear

And today we will just finish our lecture at genetic elements and chromosome this is the last slide of today's lecture and then tomorrow we will talk more in detail. So, for today I want to just make sure that you understand that these are very strong bond between A T between the 2 strands of double stranded DNA, but they can be removed by denaturation and everything else here I have already mentioned.

So, here we have a summary of the genetic elements that I talked about earlier in the class. So, chromosome double stranded DNA. So, it means it has 2 strands and usually found in prokaryotes like prokaryotes is where from. So, also very relevant they extremely long and when they found in prokaryotes they have usually circular. So, prokaryotic chromosome will be circular. So, there will be no singular end unlike if chromosome is found in eukaryote it will be linear still double stranded, but linear now plasmid is the extra chromosomal genetic material which is usually double stranded can be circular can be linear it is found in all, but it is less common in eukaryotes and this is extra chromosomal is an it is not part of the chromosome.

They are usually shorter than chromosome much shorter can be circular or linear then we had transposable elements they are found in all these here also extra chromosomal sorry these are also genetic material, the difference between plasmid and transposable element is that transposable genetic element you will enter the chromosome unlike plasmid which stays away from chromosome. And then we have organellar genome and organellar genome is very relevant in eukaryotes.

So, in eukaryotes remember we have organelles such as mitochondria and chloroplasts now mitochondria and chloroplasts have their own genetic material, which resembles the generating material of prokaryotes this is known as organellar organellar genome and this is usually medium length compared to eukaryotic genetic material.

And circular and this is a similarity it shares with prokaryotes and then we have virus which can be single stranded or double stranded DNA or RNA they are usually shorter circular or linear there is lot of diversity in viral genomes. So, my dear friends this is all for today in next lecture we will go ahead and revise briefly go through what we had talked today and proceed forward and dig into that more details of the central dogma of biology.

Thank you very much.