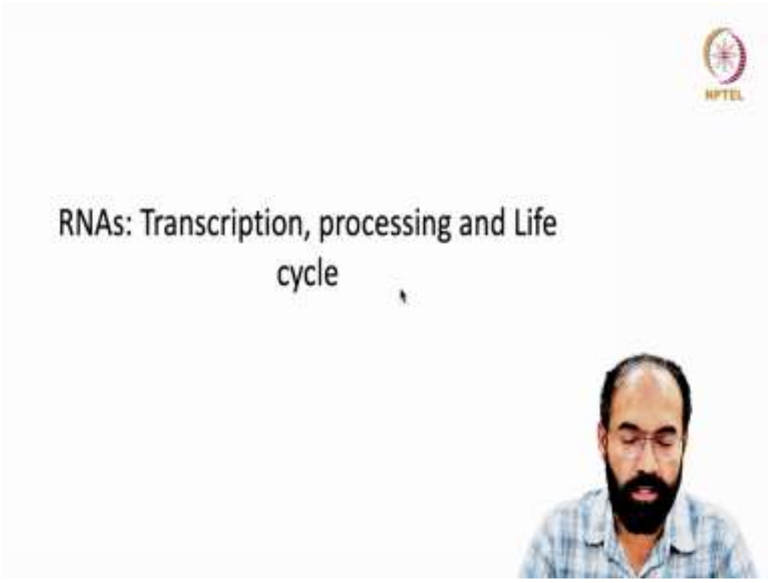
RNA Biology
Prof. Rajesh Ramachandran
Department of Biological Sciences
Indian Institute of Science Education and Research, Mohali

Lecture - 10
RNA Transcription: The Central Dogma

(Refer Slide Time: 00:21)

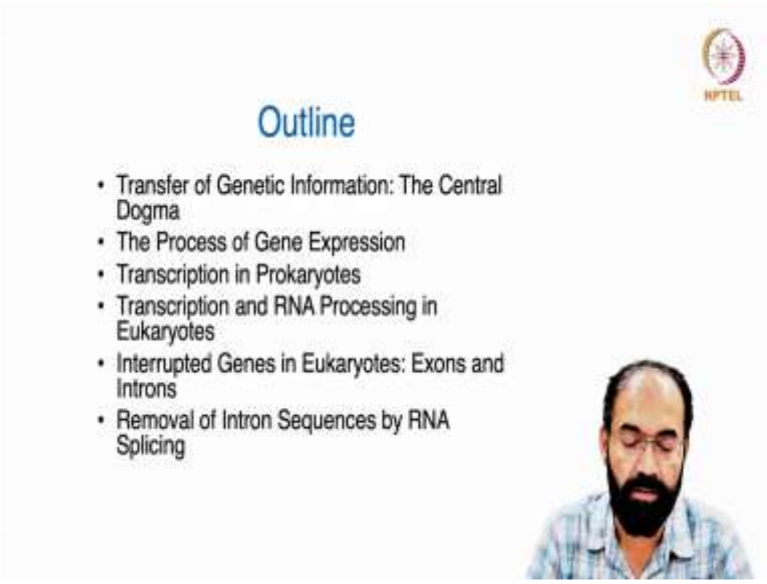


Welcome back to another topic of RNA biology; so, today we will start with a new section which talks about the RNAs, its transcription, its processing, and the life cycle

(Refer Slide Time: 00:38)

So, this is a structure of the RNA polymerase which is a protein enzyme which helps in the production of the RNA from the DNA template. It is having the shape of an inverted C and it basically clinches on to the DNA strand. Remember whenever the transcription takes place only one strand of the DNA act as the template. During DNA replication both the strands act as the template; whereas, when DNA contributes to the production of the RNA only one strand of the DNA act as the template.

(Refer Slide Time: 01:18)



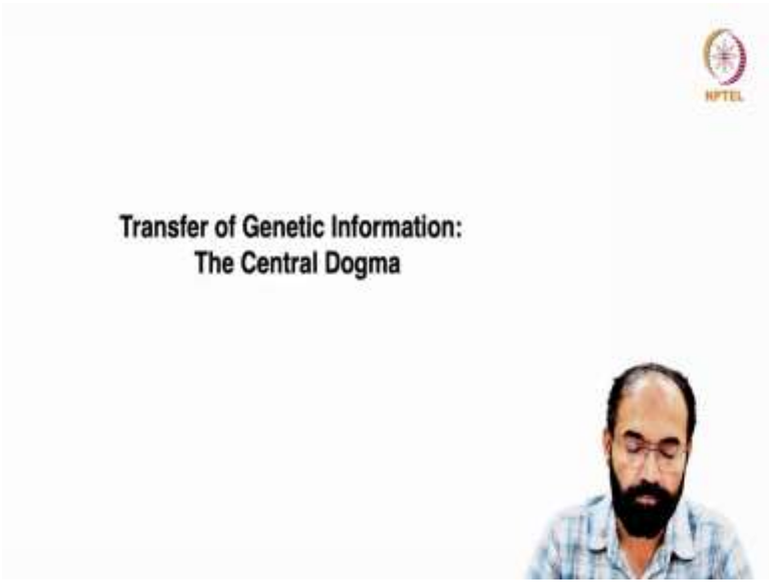
And in this topic, we will cover a few topics as listed here it is an outline. So, transfer of the genetic information that we call it as the central dogma. And central dogma basically means that a routine step of functioning is always followed, no matter which organism you are talking about. If it has a genetic material, it has to follow a routine procedure we will see more in detail about that.

And then the process of gene expression how the so called gene expression takes place in a cell and then transcription in prokaryotes, prokaryotes are bacteria and other single celled organisms and who does not have a proper nucleus that is what we refer to as prokaryotes whereas, the next topic is the transcription and RNA processing in eukaryotes; that means, those organisms that have got a proper nucleus.

Every organism that have got a nucleus is a higher order organism which has got a higher level of or a higher complexity in terms of its gene expression and functioning. And then we also learned about Interrupted Genes in Eukaryotes, interrupted genes basically means in a gene or a sequence there are wanted as well as unwanted genes unwanted regions.

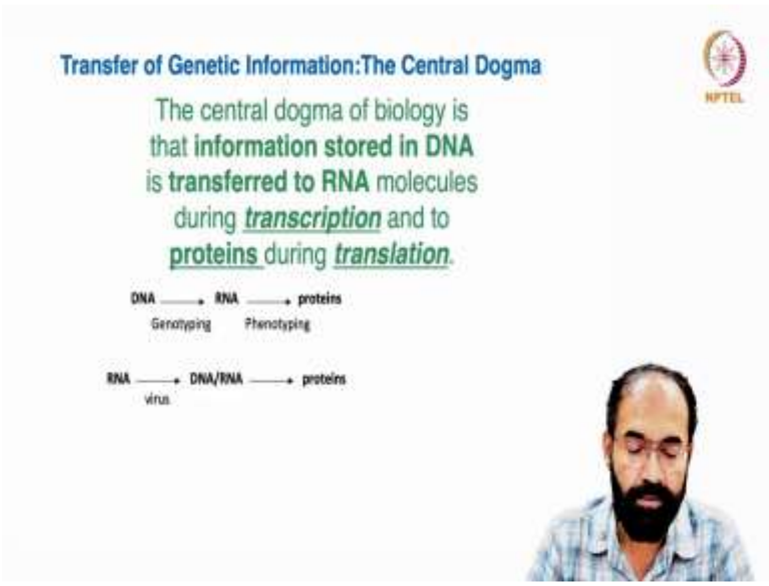
So, we call those genes as interrupted genes, means in order for these genes to function we need to get rid of this unwanted portion, unwanted area should be removed before this RNA can function as a proper mRNA for the translation. And we also we learn about the removal of the intron sequence by the process called RNA splicing.

(Refer Slide Time: 03:06)



And the Transfer of Genetic Information, the Central Dogma in molecular biology.

(Refer Slide Time: 03:13)



What it basically means is the information is stored in the DNA. We have already learned that in the RNA world hypothesis the information was generated randomly in an RNA which is now getting converted into the DNA for the safekeeping purpose. And so, this information stored in the DNA should be made available as and when required; so, that process has to be done through RNA transcription.

So, the process of formation of RNA from the DNA we call it as RNA transcription and this transcribed RNA now gets converted into protein through a process called translation. And so, that is what mentioned here DNA getting converted to RNA and RNA getting converted to protein.

So, the DNA to RNA or the information present in the DNA, a character present in the DNA is basically called as the genotype of an organism. And this RNA when it give rise to protein or because, the proteins are the one which is going to perform a task they are the workforces in a cell; so, that functioning is referred to as phenotyping.

So, genotyping and phenotyping can be explained in a simplistic term. Say for example, if you know to draw an artwork or you know a you know how to sing you are a classical singer. But whether you sing or whether you perform your artwork depends on the circumstance. So, genotype is what? The ability to sing or ability to perform an artwork is the genotype. Phenotype is the one whether or not you will sing or whether or not you will paint; so, that is what phenotype.

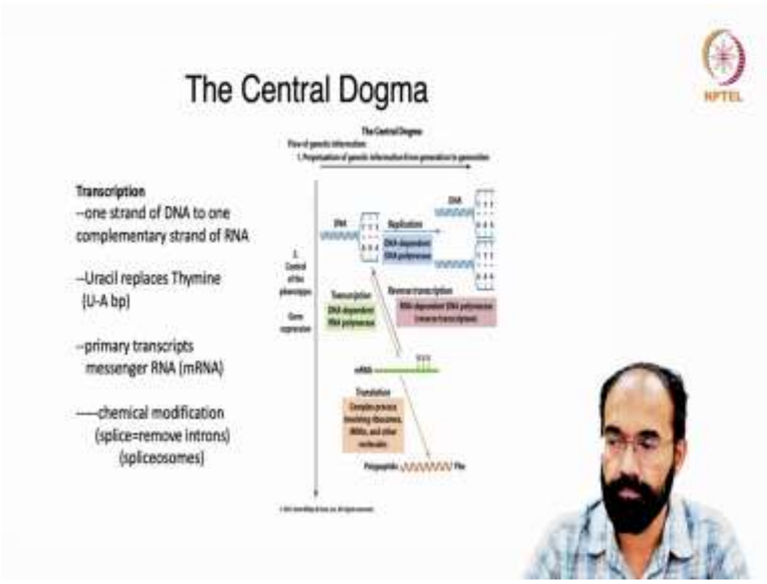
So, genotype is there does not mean that it has to express the phenotype. Say for example, in your fingertip the cell any given cell if you take from your fingertip or a on the root of your hair has the entire genome of you entire information required for an organism starting from its development throughout the functioning is present. Roughly we have around a 23000 genes a human being has. All the genes are there, but they are not expressed, any given time around 10 to 12000 genes are expressed in a given cell.

So, what it indicates that the number of genes that is the genotype need not necessarily express any given time; based on demand, based on situation this happened. And let us think about situation in an RNA in an RNA virus. So, RNA viruses are plenty you know COVID virus, influenza virus, many more like even HIV virus many are RNA genome viruses; so, they do not have DNA in them.

But what they do they either make a copy of itself that is an RNA copy produced from the RNA itself or RNA gets converted into DNA depending upon the nature of the virus, this is how they fall into the central dogma. In any case once the RNA getting converted into DNA, it has to undergo a process of transcription again in order to have the phenotype of the virus to be expressed.

Or say if an RNA is making a copy of another RNA, then it can participate in the protein translation directly, depending upon which virus you are talking about and which host which organism we are referring to this may vary. But why we mentioned about virus here is, because the central dogma has to be followed strictly and it is unchallenged.

(Refer Slide Time: 07:04)



And now, when you talk about the central dogma, we can see the transcription takes place from only one strand of the DNA and that we call it as template strand. And normally a DNA strand when it is present it is double stranded and they are complementary to each other. Say in one strand you have got A, then the opposite strand you have got a T that is thymine A stands for Adenine, T stands for Thiamine.

If one strand has got G then the opposite strand has C; so, what we have to say they are not identical copy, but they are complementary copy following this route, A pairs with a T and G pairs with a C, like that it continues; so, that is the double stranded DNAs quality. Same way if one strand is of the DNA is acting as a template, the A containing region.

Now, have an RNA which has got to U instead, Uracil instead of thymine, because we know thymine is a feature of DNA, thymine is not present in the RNA. But whereas, if a G is present in the template DNA, then there will be a C, C stands for Cytosine, G stands for Guanosine.

So, this complementarity rule is followed when a RNA is transcribed from one DNA strand or one DNA template. And this give rise to primary transcript and we call it as messenger RNA. And this RNA has to undergo some chemical modification depending upon the organism, some some process are called five prime capping and three prime polyadenylation.

And then sometimes if a eukaryote have got interspersing, region such as introns if they are present they have to be removed a process called splicing mRNA splicing. In and this is helped by structures called spliceosome, without which you will not be able to make a mature mRNA and that is what you are seeing here in this right-hand side panel.

You can say that this DNA when it is opened up, it is capable of making a copy of itself through a process called DNA replication, for that it use an enzyme, DNA dependent, DNA polymerase. And now, once it is copied each strand participate in the production; so, one double stranded DNA now end up getting two double stranded DNA, because one cell become two two become four like that.

So, DNA dependent DNA polymerase helps in the production of DNA copy. Now, another enzyme comes into picture, remember these are all protein enzyme that enzyme is DNA dependent RNA polymerase. So, name itself indicates, it needed DNA to act as a template and the product is RNA. So, DNA dependent RNA polymerase comes into picture and it make a copy of RNA or it transcribes an RNA by making use of one strand of the DNA.

And this mRNA if it is processed properly, it can participate in translation. And this is a complex process that involves transfer RNA, ribosome's and many other protein factors for the proper termination of the translation etcetera. We will see about that more in detail, but end of the day or end of this process of translation you end up getting a polypeptide. It can be an enzyme or it can be a structural protein, it can be anything because remember the proteins are the major workforces in a cell.

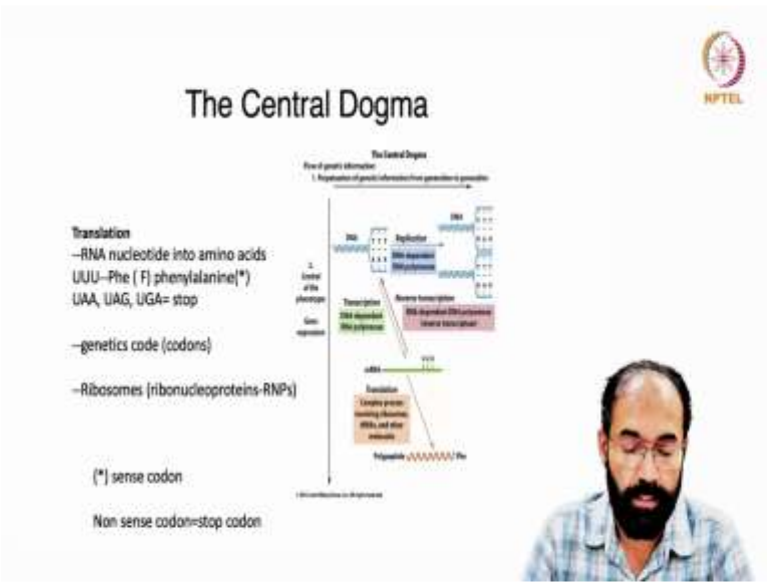
And now, there are some viruses we discussed that they have RNA as their genome; say for example, HIV Human Immunodeficiency Virus which causes AIDS in human which is an RNA virus. So, this RNA virus make use of an enzyme that is called reverse transcriptase, transcription you know DNA to RNA is called transcription. What is reverse transcription? Just opposite of that RNA becomes DNA.

And this DNA can now integrate into the host genome and become part of the host genome and such viruses are called retroviruses. Because, they enable their RNA genome to be converted into DNA and they deceive our system that fool our system it gets integrated and our body cannot recognize which is our body DNA or which is the viral DNA.

So, it will keep propagating along with the cell; and as and when this virus required to transcribe itself, it will make a copy of the RNA from this integrated genome and it will propagate itself. That is why retroviruses once they are integrated it is very hard to very very difficult to destroy. So, we are talking about four different enzyme; one is DNA dependent DNA polymerase, and DNA dependent RNA polymerase.

And there are enzyme which can make a copy of RNA from an RNA that is RNA dependent RNA polymerase and then comes reverse transcriptase that is making a DNA from the RNA. So, all four possible enzymes contribute some way or the other in the process of called reverse in the process of called central dogma.

(Refer Slide Time: 12:55)



And in the translation when you see, translation is basically conversion of the RNA nucleotides into amino acid sequence. Now, how does it do? As you can see here there are three U's are mentioned here UUU which is present in the RNA which act as a codon and this brings in phenyl alanine. Phenyl alanine is an amino acid that is attached on to a tRNS.

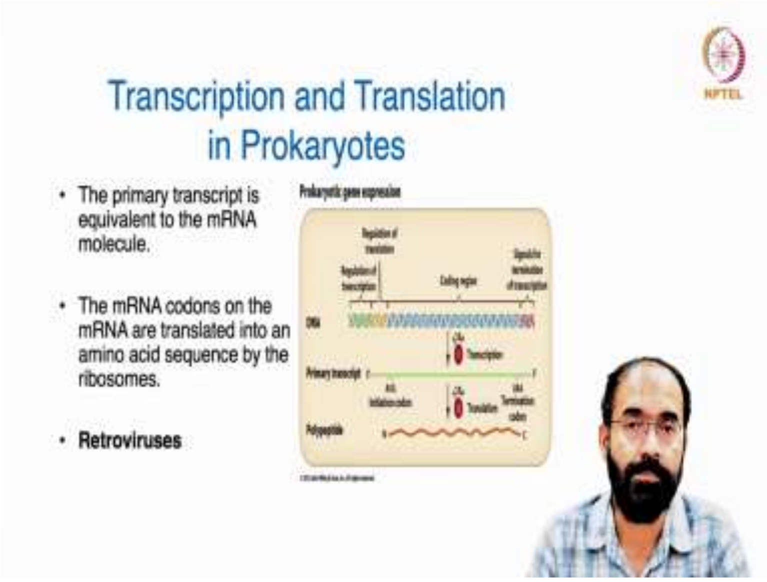
So, that tRNA has got anticodon that can pair with this UUU; that means, the anticodon of the phenyl alanine bearing tRNA will have AAA triple A will be there and during protein synthesis that will come into picture. And then you also have three stop codons, only three stop codons are there starting from bacteria to human all organisms have got only three stop codons. One is UAA, UAG, UGA they also have names called ochre, amber, opal like that and they do not bring in an amino acid and hence we call it as stop codon.

So, wherever there is any of these three codons are there you will end up getting a stop codon and total 61 meaningful codons are there and these three dead stop codons are also there. And total 64 codons are possible and we will see the codon preference table etcetera in or the genetic code etcetera in the subsequent classes.

So, basically these triple bases are called codons and every amino acid will have to have three bases. Or in other words if you have an RNA that has a coding region of 300 bases it can bring in 100 amino acid, why? Every three base brings in one amino acid. And for doing this you need to have ribosomes which are nothing but ribonuclear proteins.

And wherever an amino acid can come in we call that codon as a sense codon and where no amino acids can come in we call it as stop codon. In the right hand side panel that is what you are seeing the same what you saw as the part of the central dogma you can see the second part is nothing but the protein translation that what we saw so far.

(Refer Slide Time: 15:26)



And if you look into prokaryotes in the bacteria, the transcription is quite simple compared to a eukaryote, because you have a primary transcript that is to be formed from a given stretch of DNA. So, you can see that this is a DNA double stranded DNA double helix what you are having here. And what is shown in this green colour is the regulators of transcription; that means, it is also DNA sequence, but it do not code for an mRNA.

It has region or area that can help in the recognition of the certain protein or recognition of some elements that will allow where to start the transcription. And then you have a yellow colour region that is the regulation of the translation; that means, it forms under some of the UTR untranslated region etcetera and that can influence whether how effectively a mRNA should be translated.

Then in the blue colour you have the actual coding region; that means, it brings in the amino acid. And then lastly you have in this the pink colour the signals for the termination of transcription. So, what do we understand? There should be signal for where to start the transcription and what are the area to be transcribed and where to stop the transcription.

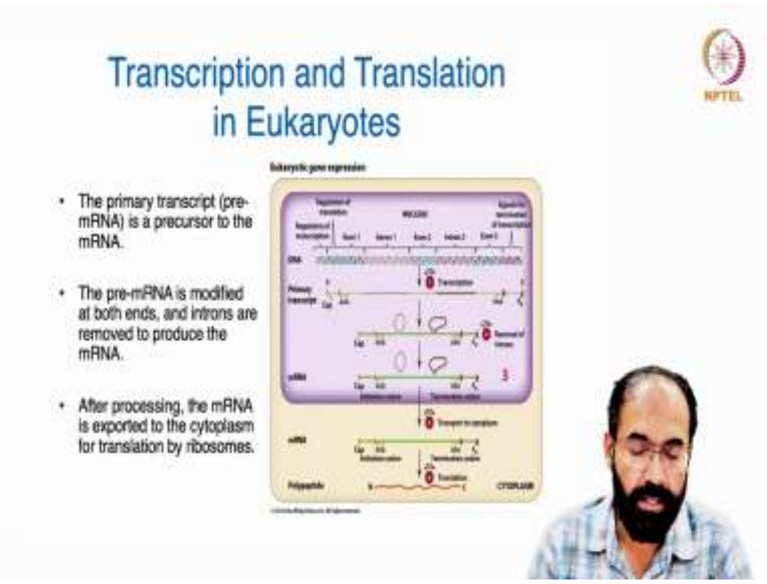
And that is the first step of transcription in prokaryotes such as bacteria is the production of a primary transcript. And since the bacterial genes are less complex, you end up getting a straightforward functional mRNA. And every mRNA will have a start codon that is AUG, AUG brings in methionine.

Every protein will have methionine as the first amino acid, no matter whether it stays back or not because in some cases this methionine will be selectively removed. But at the time of production every protein will have methionine as the first amino acid. So, start codon is only one that is AUG in the genomic sequence it will be ATG, if you are talking to a DNA it will be ATG.

So, now the second process is the production of the protein that is protein translation. So, the mRNA codons on the mRNA are translated into an amino acid sequence with the help of ribosomes and tRNA and this mRNA is now getting converted, getting converted means getting encoded into a protein sequence or an amino acid sequence.

And same procedure is followed by retroviruses, the only difference is they have to convert into RNA to DNA and this DNA is now getting stored in the host. And whenever the host is undergoing transcription, this RNA comes out, this is how it works in the retroviruses.

(Refer Slide Time: 18:34)



Now, if you look into transcription and translation in eukaryotes, it is very much similar to that of very much similar to that of the prokaryotes, the only difference is that they have a nucleus. The nucleus is pretty strong to protect the genome, pretty strong in the sense it is not available for the cytoplasmic proteins to interact or intervene.

So, the primary transcript is produced as in the case of bacteria from the DNA itself, as you can see here there is a double stranded DNA. So, the primary RNA has to be modified on both the ends. It has to undergo modifications and the introns are removed the intervening regions also transcribed, they have to be removed for the actual functioning of the mRNA, otherwise that mRNA is useless.

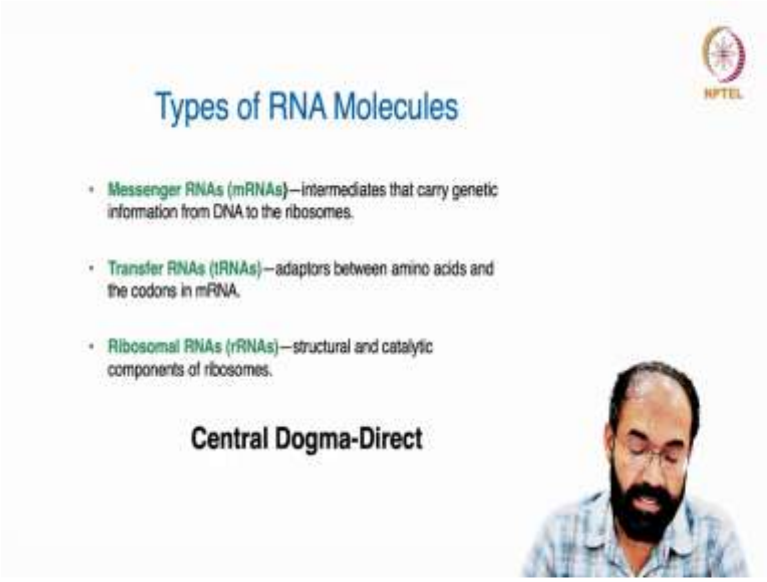
So, after processing what happens, the mRNA is now exported to the cytoplasm for the translation by the ribosomes. So, let us see here you can see there is a double stranded DNA here and it has got regulatory sequence as in the case of bacteria. So, the regulation of transcription and also elements required for regulation of translation also present in the untranslated region part.

In a mRNA it has got a coding part, and also it has got 5 prime untranslated region, and 3 prime untranslated region. Untranslated itself indicates that it is not translated it is not getting coded for protein. So, you have exon 1, exon 2, exon 3 in between you have intron 1, intron 2 which is present in between these exons. And when they are transcribed both introns and exons are transcribed.

Now, it has to be matured, it has to undergo a process and this process is called RNA splicing. The RNA splicing allows the joining of the exon and removal of the intron. And once the RNA pre mRNA is spliced what happens? The 5 prime end and the 3 prime end has to undergo some maturation process; 5 prime capping, 3 prime, polyadenylation etcetera. Once all these process happens successfully, then it will be exported to the cytoplasm.

Cytoplasm is the place where the mRNA will find the tRNA, ribosomes etcetera, and they help or they work together to make a polypeptide in the cytoplasm. So, this is what you should understand about the transcription and translation of prokaryotes and eukaryotes in a simplistic manner.

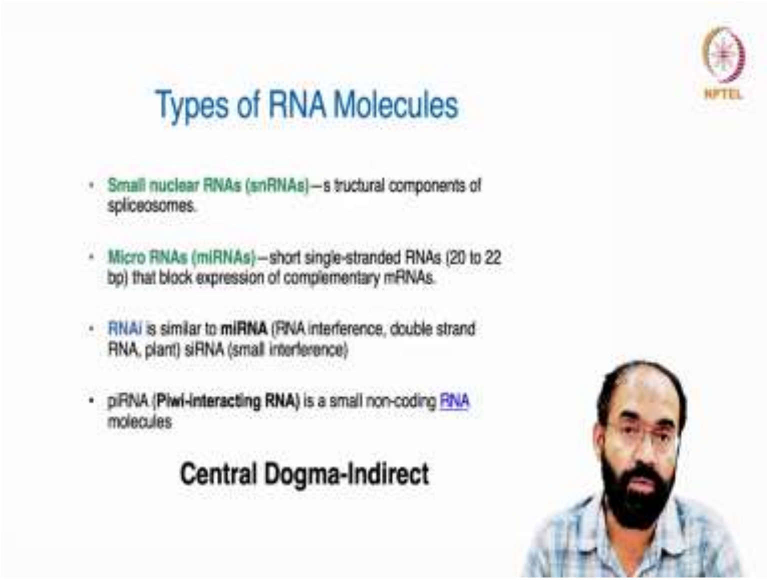
(Refer Slide Time: 21:45)



Now, let us see what are the types of RNA molecules that are present? So, types of RNA molecules they are messenger RNA that act as the messenger or a message that converts the genetic information into protein; so, they stand in between. And then transfer RNA, they are the tRNAs they help in the production of proteins and then ribosomal RNA they also help in the production of proteins.

So, mRNAs are the intermediates that carry genetic information from the DNA to ribosome’s, transfer RNA or the tRNA they act as the adapters between amino acids and codon's of the mRNA. And then ribosomal RNA they are basically structural and they also play catalytic components of the ribosome’s, this is called central dogma direct.

(Refer Slide Time: 22:49)



Now, let us see what are the other types of RNA molecules that will participate in the indirect way of influencing the production of protein. They are small nuclear RNA, snRNA they are structural components of the spliceosomes.

And then micro RNA they are coming as the short single stranded RNAs around 20 to 22 nucleotide in length that block the expression of complementary RNAs and then RNAi that is RNA interference that is similar to micro RNA, and the double stranded RNA that is seen in plant also influence the translatability of an mRNA.

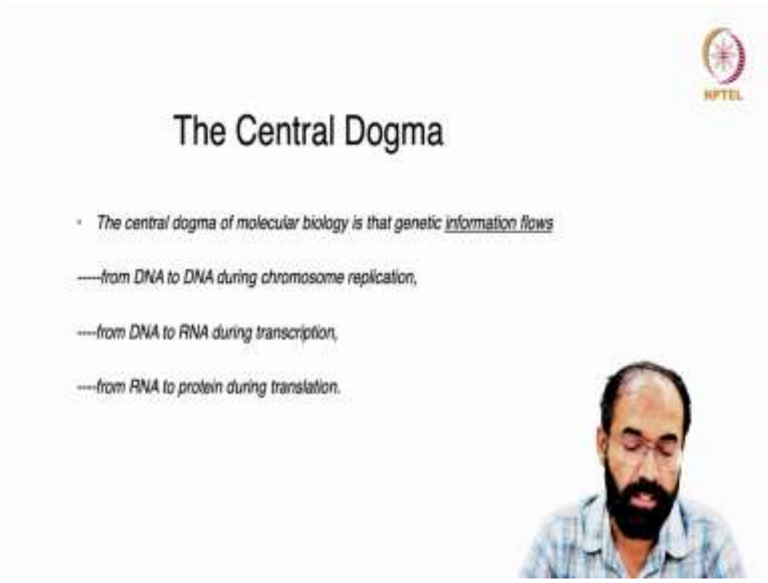
And siRNA is small interference RNA that also can influence the expressivity of an mRNA. Then comes piRNA that is Piwi interacting RNA normally seen in the germ cells identified in mouse germ cells and it is also a non coding RNA. So, what do we understand these RNA molecules are not influencing the central dogma as such directly, but it can affect indirectly.

That means, whether you have food at home or not that indicates whether you are rich enough to have food. But whether you are able to eat or are you willing to eat depends on your willingness to eat plus your body’s nutritional status is dependent on by whether or not you eat, that is an indirect way of regulating your nutritional or your health status.

So, these molecules do not influence the transcription or they are not able to influence the gene expression in the first step. But they can influence the gene expression in the second step that is the translation. So, an mRNA is there, but whether it should be allowed to express into protein or not is decided by these molecules.

So, that is why we call these molecules as central dogma indirect; that means, they are not going to affect the central dogma direct, but they can influence the central dogma indirectly.

(Refer Slide Time: 25:26)



So, so let us look quickly the central dogma once again. The central dogma in molecular biology is that the genetic information flows from the DNA to DNA; that means, DNA has to make a copy of itself using DNA dependent DNA polymerase otherwise it will be lost. And during the cell replication or the chromosome replication, and then from DNA to RNA during transcription because without which gene expression cannot happen this is also part of central dogma, then from RNA to protein during translation.

So, these three steps are the major focus points of the central dogma. All other features that will tweak the system based on which cell or which organism what condition the cell is living etcetera. Accordingly, the central dogma will be tweaked we can study more about the central dogma in the next class.

Thank you.