RNA Biology
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Lecture - 54
Telomere, Telomerase and Impact on Genomes: Telomerase Length as Marker of Aging

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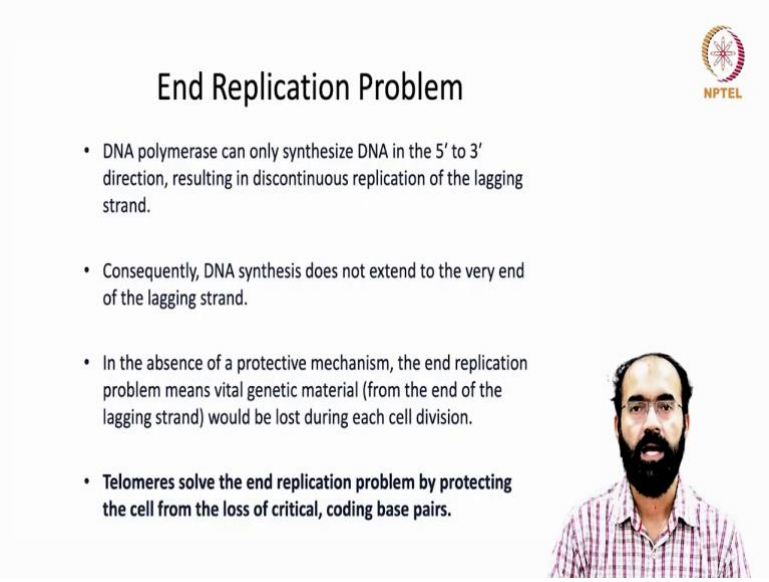

Hello everyone, welcome back to another session of RNA Biology. So, we stopped here that is the Telomere and Telomerase importance is simply because of the end replication problem. And end replication problem is inevitable in the case of lagging strand because the polymerase need a place to hold itself onto it onto the template strand and where it is holding it cannot synthesize.

So, it is a fact, but the nature of a cell or a chromosome is such that it can accommodate this end replication problem if it has the telomeres attached to the end. So, every time you are not losing gene, but you are losing telomeric repeats. So, this way this problem can be solved.

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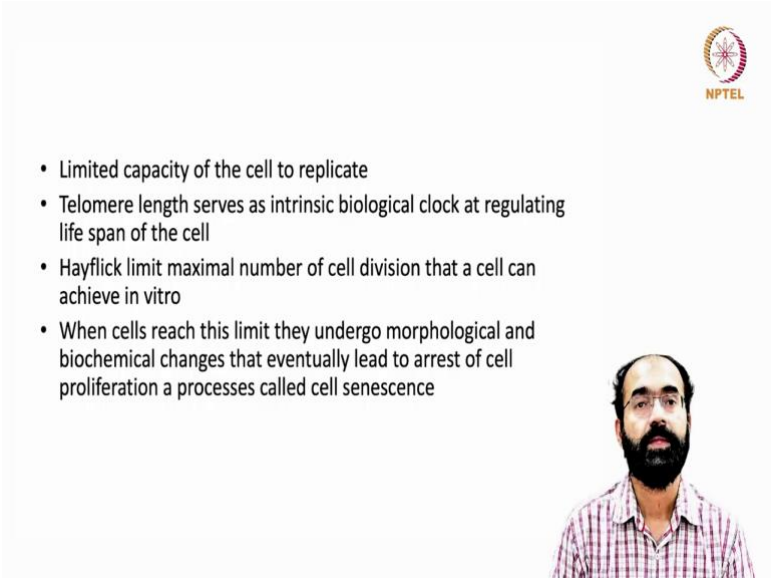

So, limited capacity of the cell to replicate is a fact credit goes to absence of telomerase in a any given cell and presence of end replication problem. These two together thing adds it up just like you have a fixed amount of money in your wallet and no one is earning money to add into the wallet on the other hand you are using it also.

End result anyone can predict. End result is a empty wallet. Same logic applies to telomere length and the senescence. So, once the wallet is empty you have no way of using money unless you find a way of introducing money. So, Telomere length serves as a intrinsic biological clock at regulating lifespan of the cell.

So, if say your body has got 100 different types of tissues or cell types you have, all of them are not dividing at same rate depending upon which tissue you are talking about they have a moderate deep cell division or non division at all like brain cells. Brain cells many neurons in your body are fixed right from your birth and they will die along with you they are not going to divide they are not going to die either.

Their telomeric point of view their length is perfect, but still they can store signs of old age credit goes to you know their peroxisomes, and other organic toxins basket their waste baskets will get accumulated with lots of you know compounds which needs processing.

So, normally the cell will process them once they divide. So, in after several you know decades the neurons also will accumulate toxins and they can die nothing to do with the shortening of telomeres because they do not divide, but they can die due to some other reason.

So, cells have different ways of dying, but telomere length is one of the important criteria for a normal cells to die. So, limited capacity of cells to replicate is a fact. So, telomere length serves as a intrinsic biological clock regulating the lifespan of the cell that is also a fact. Hayflick limit is the maximum number of cell division that a cell can achieve in vitro. You take a cell you took a cell from an organism and you are making it to divide and divide and divide without introducing telomerase. So, how long it can divide and then it will stop dividing. So, this is the Hayflick limit.

So, when cell reach this limit, they undergo morphological and biochemical changes that eventually lead to arrest of cell proliferation, a process that the cell will undergoing senescence. So, the arrest of cell proliferation. Cell proliferation is no more occurring or no more allowed then it is a process that is called cellular senescence. Cell is going to a stagnated state. So, that is called cellular senescence.

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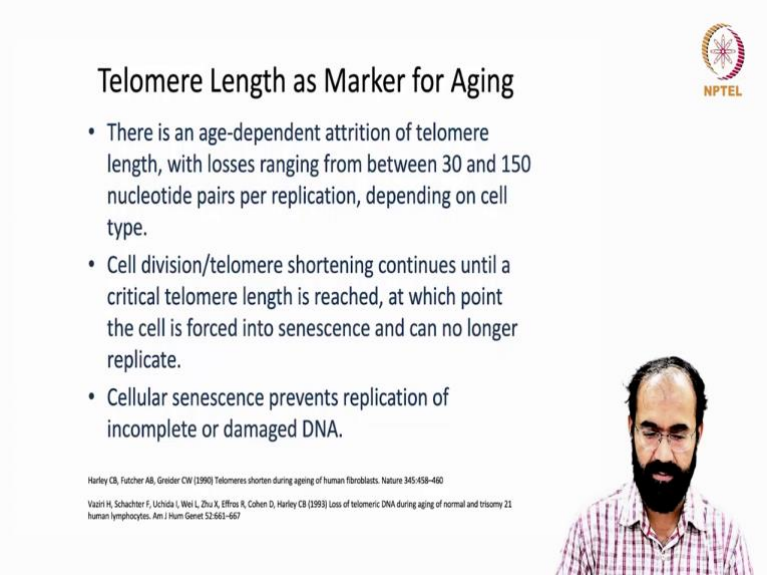

So, telomere length can be measured as a marker of aging. Like I told you when we studied about the bar bodies and the it is in activation if I give you a tissue and I am not saying whether it is from a male or a female. The best way you can check is make a

smear of that smell that cell or tissue and put it in a glass slide and observe under a microscope.

The nucleus will have a distinctive dot which is absent in males, but present in females. Any organism like irrespective of which is mammal species you are talking about they will have a dot and that is the bar body. So, you can identify. Same way you take a cell and measure the telomeric length you can clearly tell how old this cell is or how old this cell could have come from a person.

So, if you have a tissue given you all you have a set tissue like for forensic studies etcetera. People can also identify age of a person from the bonds and telomeric length also is an indicator. Average indicator we cannot say precisely if a person is 43 years old your assumption may say that he is 45 or maybe he is in 40s. No one may be able to precisely say he is 43 and 6 months you cannot say, but it will give you a clear indication.

So, cell division can cause the telomere shortening and it will continue until the critical telomeric length is reached at which point the cell is forced into senescence and can no longer replicate. That is what is important it is now doing nothing, it is deciding not to divide, it is in a stagnated state. And if a cell is in a stagnated state in spite of there is a need for dividing then that cell will eventually will be marked for apoptosis.

Because that is not something is required in a normal organism means the system recognize this cell is not up to the mark this cell is not doing. That is why in some people human beings very old people like who are 95 or 98 or even 100 plus some people you will see their skin will be peeling off just like that, their skin is not very tight.

Their so, it is a tissue damage basically they do not have its not any autoimmune disease or something, very old people their skin will be so weak that it can simply peel off like a onion skin. So, it is not holding on to the place properly. And some people it is so serious that it can look like you know real injury wound no one is peeling it, but skin is not sitting in the place.

So, that can invite formation of wounds etcetera. So, it is simply because the cells are very old. So, they are not dividing the rate at which it should have been dividing in their

young age. So, so cellular senescence prevents replication of incomplete or damaged DNA.

And this senescence or stagnation is a necessity because if it continues to divide say like your monthly income is 1000 rupee and your monthly expenditure is 5000 rupee you can imagine where that person will end up. Either he should be become an antisocial element or he should be borrowing money. How long you will leave with borrowing money? So, if your 1000 rupees your income, your expenditure cannot exceed 1000 rupee, it can maximum go is 1000 rupee, if it goes beyond that then that is a problem.

Same way if the telomeric length is shortened if it still continue to divide, then what will happen? You will have gene loss. And if gene loss is their cell will not be able to perform the task and it can also invite mad behavior of the cell and it can lead to cancer. Mad behavior means cells have to follow certain rules that contact inhibition, do not pile over each other or when one cell comes in contact with another it should stop dividing etcetera.

So, you do not want such things to happen in a normal scenario, but cancer cells do not follow any of these rules they continue to divide continue to divide pile over each other. So, such and they do not require any anchorage, they do not need any substratum to bind etcetera. So, all these things can lead to very serious complications if a cell decided to continue to divide. So, you do not want that to happen. So, that is why stagnation or senescence becomes a bliss for the tissue to avoid becoming cancerous.

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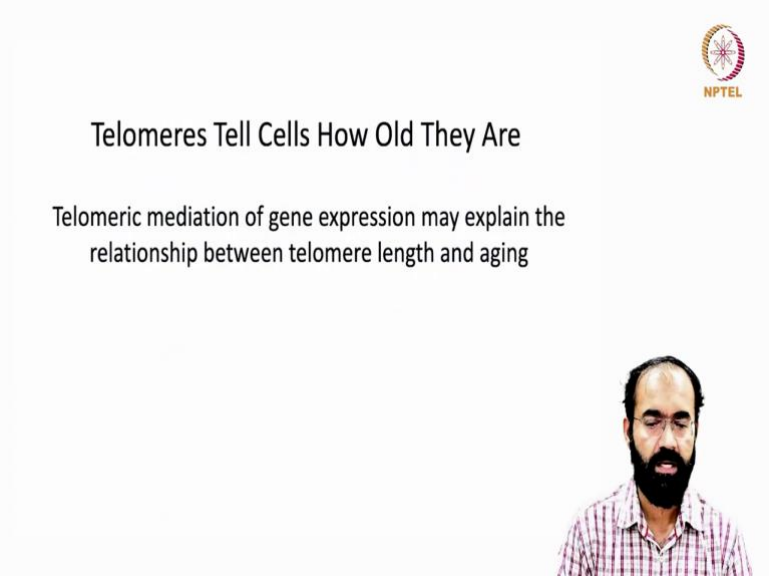

So, telomeres tell the cells how to how old they are or how to respond to a shortened telomere. So, telomere is basically a supervisor you can call it telomere as a supervisor. And you should also understand that telomere have got specific protein telomere binding proteins. So, if a given cell have got telomere length such that 100 protein molecules can bind let us assume, 100 protein molecule can bind and any given cell will continue to produce that many proteins.

So, if the telomere length is ok, all the 100 cells will be able to bind down to them, but if the telomere length is shortened only 90 cells are 90 proteins are only able to bind remaining 10 is sitting jobless. So, they will trigger a senescence reaction, they will go and complain to the system that you know do not divide because we are sitting jobless. They just like if you want to enter your house and door is not opening.

So, someone has locked from inside, you will not say ok, I will keep sleep outside. You will alert people either neighbour's, police etcetera, say something is wrong, my door is not it is locked from inside, no one is opening. Same way these proteins will alert the system that we are not able to bind. So, let us kill this cell or stop further dividing because we are sitting jobless that is an indicator.

So, you may wonder who is there monitoring with a microscope or something inside the cell to measure the length of the telomere. So, this is how these proteins will alert the system that telomere length is short. So, I will not go into the details of all these proteins

because there are bunch of proteins are there because our focus is to understand the telomere length and how they are maintained.

So, telomeric mediation of gene expression may explain the relationship between telomere length and age because they can influence the gene expression events also.

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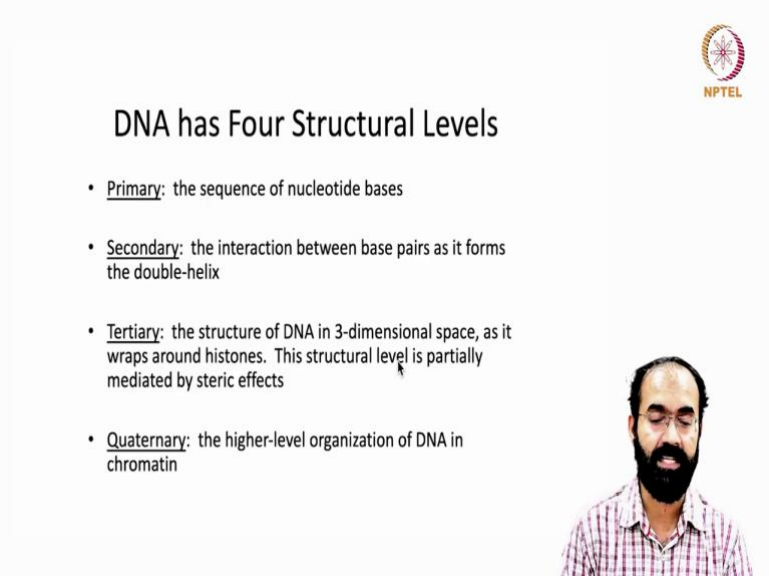

Let us see with some example. DNA basically have got four structural levels. First one is primary that is the sequence of the nucleotide itself. Secondary structure basically the interaction between base pairs as it forms the double helix.

RNA also we have seen primary structure and secondary structure then comes a tertiary structure. The structure of DNA in three dimensional space that means, how they are folding though over the interaction of different domains of the secondary structure as it wraps around the histones.

So, DNA the three dimensional structure is through histones which forms the chromatic structure and it compacts we have seen this in the x chromosome inactivation in detail. So, this structural level is partially mediated by steric effects. So, this sequence of telomere or other non-telomeric regions they have a distinct way of compaction of the chromosome. So, then they also have a next level of structure called quaternary structure that is the higher level organization of DNA that is giving rise to chromatin.

So, DNA sequence is the primary and its interaction with other DNA strands is secondary and the tertiary structure is bringing in contact with the histone proteins and quaternary structure is how this histone protein and the DNA interact with each other and form a much compact structure called quaternary structure.

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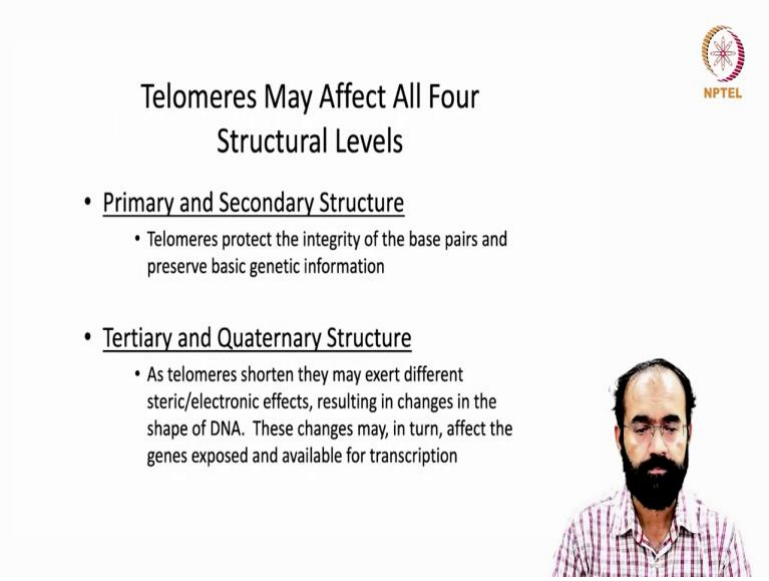

So, telomeres may affect all our structural levels. So, they can influence all this structure because primary starts with the sequence itself that is T 2 Hg 3. So, primary and secondary structure can be influenced by the telomeres and they protect the integrity of the base pairs and preserve the genetic information.

Means it act like a guardian, it will preserve the integrity of the chromosome ends. It is like a solid wall just like your chappal if you buy a high quality shoes the bottom of that shoe will be very solid material to prevent erosion. And on top of that there will be a layer where is your feet is touching, that will be soft. So, if you make a shoes with that soft material within one month it will erode off.

So, there is a protection to the sole of your shoes with a very hard material, we are very resistant to erosion in good quality shoes. If you see it you will know two different types of materials will be made to put into together in the bottom of the shoes. So, in tertiary and quaternary structure as the telomeres shorten, they may exert different steric and electronic effects resulting in changes in the shape of the DNA, how the DNA is folding to in the form of a chromosomal structure.

So, these changes may in turn affect the genes exposed and available for transcription. So, the tertiary and quaternary structures and their dynamics can influence the remaining part of the chromosome.

Tertiary and quaternary structure we are talking about in the ends of the chromosome they can influence how the internal portion that is in between two telomeres is the chromosome or the genetic or the gene part of the chromosome. So, whether they are allowed to express or not also can be decided by this tweaked tertiary and quaternary structure by the telomeres.

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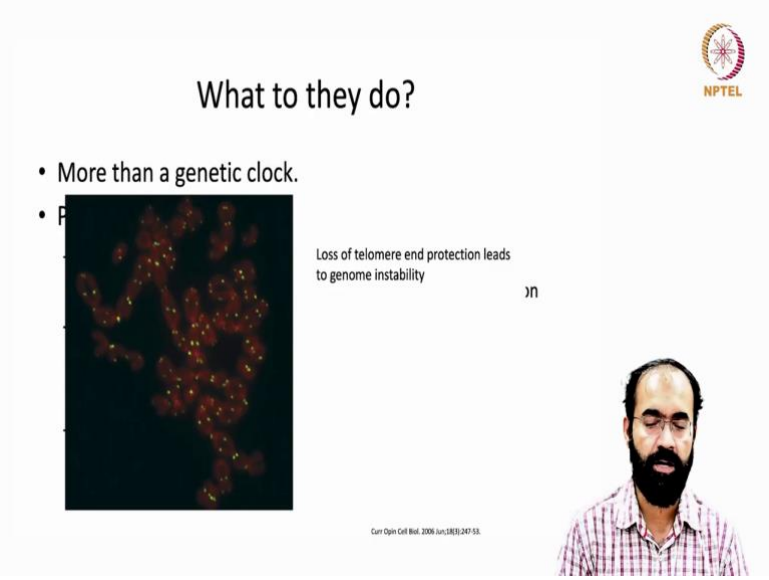

Let us see what they can do; they are more than a genetic clock. So, the loss of telomere and the protection in the absence of protection or the absence of proper telomeres lead to genetic instability. So, you do not want the ends of the chromosome not protected. Because even the telomere is contributing to the gene expression events through its ability to influence the secondary and tertiary structures.

So, telomere is not only handling the end replication problem, but they can also influence the gene expression events in the rest of the chromosome. So, you can see here this is a staining fluorescence in situ hybridization of a metaphase a chromosome where they are you can see the yellow colour dots are the telomeres.

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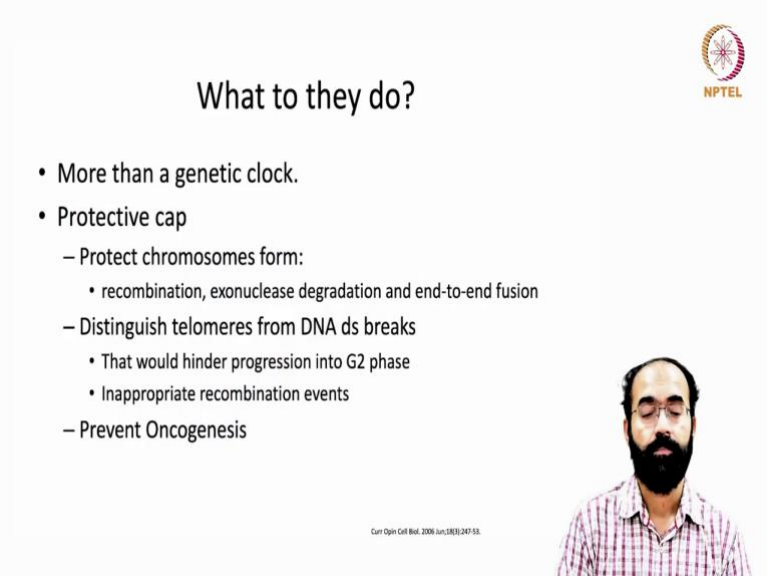

So, let us see what are the things telomere is able to do. It is a protective cap, it is a genetic clock and it protects the chromosome from recombination, exonuclease degradation and end to end fusion. So, telomere mediated secondary and tertiary and quaternary structures, it can influence or protect the chromosome from recombination means a part of the chromosome integrating elsewhere in the genome or a piece of the chromosome number 1 is now recombined into chromosome number 6 or chromosome number 10.

So, this is recombination because it went to specifically to that chromosome. Because a small stretch of sequence was similar. So, because of that it recombined during cell replication. So, because during cell replication DNA repair enzymes etcetera will be very active. And so, recombination can happen easily during DNA replication S phase.

So, it can help in distinguishing telomeres from the DNA double strand breaks. So, if there is a DNA end that is lacking the telomere, that is a indicator of broken chromosome or broken DNA. Otherwise, a DNA end is always will have a telomeric repeat.

If there is a DNA end that is lacking the telomere length or telomere sequence that indicates it is a freshly broken chromosome, freshly broken chromatin from where somewhere in the middle of the gene. So, you do not want that problem left unnoticed. So, that is why telomere is also an indicator of what is the nature of this DNA end. So,

DNA double strand break has to be distinguished that would hinder the progression into G2 phase in after a space it should move on to the G2 phase.

So, absence of this proper detection of broken DNA and it has to be repaired if there is a broken DNA, the absence of such DNA fragments present in the S phase can lead to senescence and that cell will be marked for apoptosis. If everything is perfect then the cell will move on to the G2 phase and then M phase.

So, inappropriate recombination events also will be recognized. Because telomere make sure that no chromosomes can fuse each other as long as they are present. But if a DNA is broken that fragment has to integrate onto another chromosomes end that will pause that will possibly happen if that receiving chromosome lacks the telomeric repeats.

But if that other chromosome have a telomeric end then this broken piece of chromosome cannot fuse. So, it is a kind of double edged sword. So, if broken DNA is there it has to be detected by the system and it will turn on you know DNA repair enzymes and if it fails it has to turn on apoptosis etcetera.

But what if this broken DNA immediately get recombined or fused with another region in the chromosome? Then you have a complete genomic instability. So that means, the a gene is broken and it is now situated elsewhere, that can lead to a lot of problem. So, it has to be sensed and detected.

So, telomere gives you an idea that whether such a broken piece can be highlighted effectively. Because every end of DNA in a normal cell must have telomeric repeats. So, inappropriate recombination event can be prevented and it also prevents oncogenesis. Oncogenesis is the formation of cancer, spontaneous formation of you know transformed cell. Transformed cell means misbehaving cell that do not follow the rules present in a organism and that can give rise to cancer. Normally your immune system detects it.

Normally, your apoptotic pathway comes into action, but cancer cells are smart they can find ways of evading your immune system and they can also escape apoptosis. That is how a mad cell established in an organism as cancer cell. So, telomeres also give an indication. It gives a warning symbol to the cell's surveillance system how not to allow the cell to proceed if there is a damage is detected.

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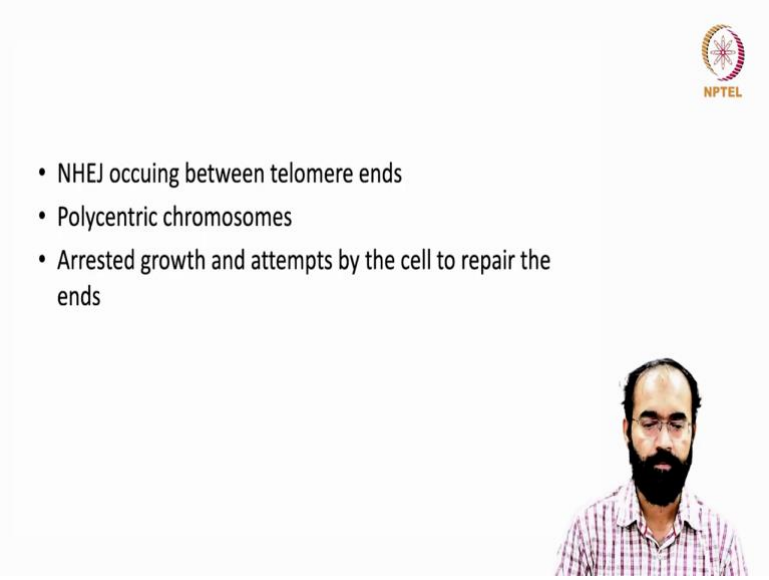

So, NHEJ; that means, Non Homologous End Joining occurring between telomeric ends. Non Homologous End Joining like if you have a stretch of DNA, if it has to recombine elsewhere, it should find some region something similar. Or in other words if one of the DNA strand is broken.

So, it will make use of the other strand, DNA has two strands know. So, if one strand is broken then it will compare look into the other strand and repair it. But if both strands are broken now you have to fuse them back. Then it will be fusing randomly and this fusion is called non homologous end joining. Means this fusion is just a fusion for the sake of saying it is a fusion.

The genes are damaged cells are damaged, but that need not necessarily lead to cancer. Say your cell your given cell have got say 10,000 genes expressing let us assume, but none of these 10,000 genes are affected because of this damage then it is ok, nothing will happen to the cell, excuse me.

If this continues to be present in a cell which is requiring this gene then what will happen? Then that particular genes function is compromised, then you will not be able to continue further. Cell is there, but it is compromised. This is an indicator that the cell can become cancerous eventually, in some case. I am not saying that one gene is not functioning in a given cell that will lead to cancer, but it gives the propensity to become

cancerous. Say in your fingertip, you have many genes required for brain development or animal development or heart formation or lung formation.

But why you need heart or lung in your fingertip? You do not need it. So, those genes are all kept off. So, if there is a deletion in those genes in your fingertip, what will happen? Nothing will happen to your fingertip; fingertip will continue to function as it is or as it was.

But if one of the genes required for the functioning of say nail formation in your fingertip nail if nail is pollen or damaged nail has to form. So, one of them is damaged, then your nail will stop growing, it will stop forever, it is because one of the crucial genes are now affected.

So, such kind of situation there is a good chance the cell can start behaving odd and it can lead to cancerous condition can. So, polycentric chromosomes can result like, if two chromosomes used together instead of one centromere every chromosome by default has only one centromere. If two are there then it will give problem when the cell division has to happen. This is a strike you have two handles for your bag and you are holding and your friend also is holding, if you are going in a synchronous manner, it is, ok.

What if your friend is walking to the north and you are walking to the south holding the bag, then what will happen? Either one of you stop moving or the bag will tear into two pieces. This is what happens when multi centric chromosomes exist. If more than one centromere is there in a chromosome, then it can lead to damage to the chromosome itself and eventually the cell will die.

So, they are arrested growth and attempts by the cell to repair the ends can result in a very serious situation that means, arrested cell growth must be marked for cell death. So, attempts will be made by the cell to repair it that there is a damage, there is a problem, but not all problems are repairable. In that situation what will happen? The cell will simply start misbehaving and it can lead to cancer.

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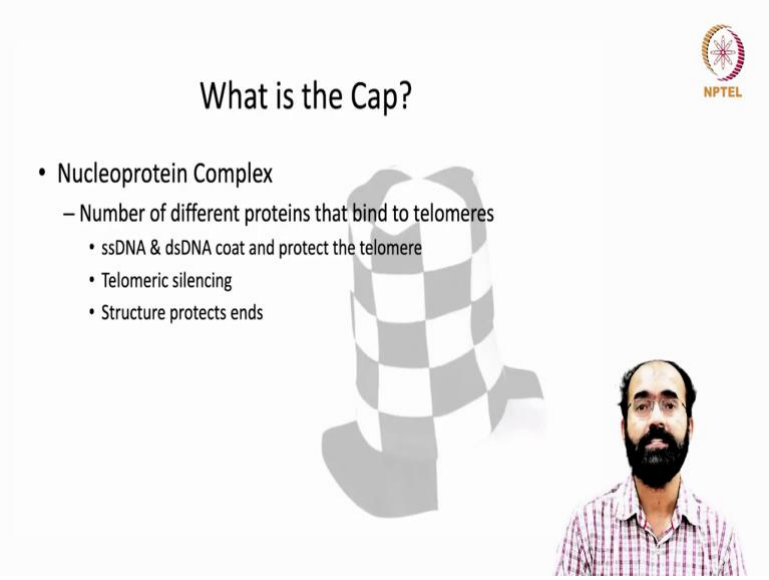

So, what is the cap? So, it is basically the telomeric cap is nothing but a nucleo protein complex. So, number of different proteins that bind to the telomeres exist. And telomeres are not typically binding to a histone protein; they have specific proteins, they are telomeric repeat binding protein.

So, single stranded DNA and double stranded DNA coat and protect the telomere. So, telomeric repeat normally can fold in such a way that it can form a complex structure with the help of protein. And it can also bring down the telomeric silencing; that means, telomeric genes once they are quoted, they remain quite dormant and exposed only during cell division. Some cells like muscles neurons etcetera they never divide.

So, their telomere is pretty strong and it will solidly protect the ends of the chromosome and prevent it from chromosomal fusion. If there is a broken DNA let it that let that cell die or that broken DNA die do not allow that broken DNA to come and attach on to another chromosome.

So, this is almost like wearing a helmet when you are driving a two wheeler or when you are going in a space mission etcetera. So, that will protect your vital organ head. So, same logic applies when it comes to the ends of the chromosome. We will learn more about cells division and telomeres in the next class.

Thank you.