Bioengineering: An Interface with Biology and Medicine
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Lecture - 26
Cell Cycle Disregulation and Cancer

Welcome to MOOC NPTEL course on Bioengineering: An Interface with Biology and Medicine. So today we are going to talk about cell cycle and in which way cell cycle dysregulation may cause diseases like cancer. We are going to talk about why mitosis is so important, why meiosis is central to reproduction. We are also going to look into various factors, internal and external factors which regulate cell cycle.

We are going to talk about the difference to the normal and cancer cells and what are these very basic molecular biology research in which may have started investigating about the cell and its properties to try to find some clues for cancer. Let us first start with cell cycle.

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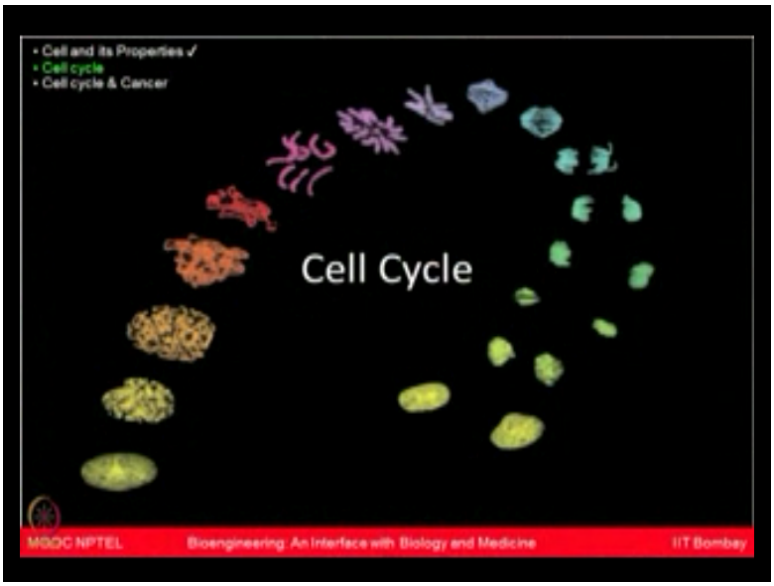

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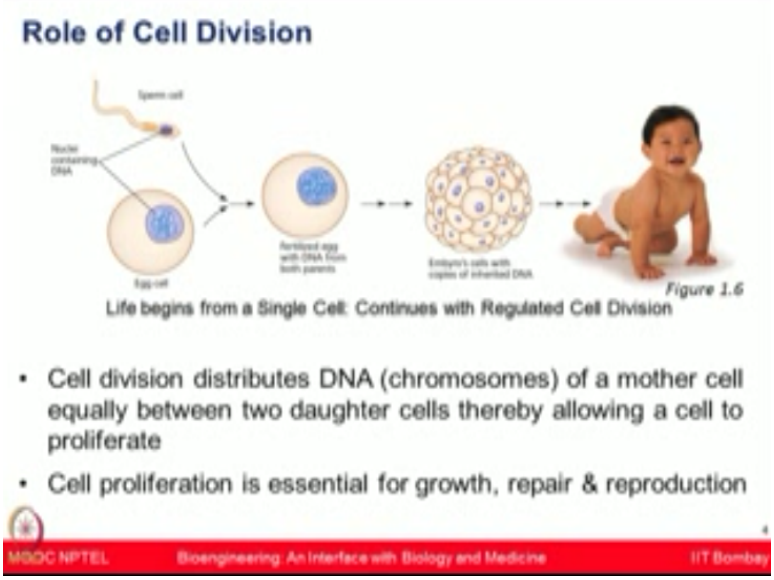

So you can see the life begins from a single cell starting from the sperm and egg cell, their fusion in the process of fertilization that gives rise to the embryo and from embryo, from the one cell.

Now after the continuous division the life starts. The cell division distributes the DNA of a mother cell equally between 2 daughter cells. Therefore, it allows a cell to proliferate. This cell proliferation is very crucial for the growth, repair and the reproduction process. So what is the cellular organization of the genetic material? So if you think about broadly a genome actually that consists of the entire DNA molecules in case of eukaryotic cells and if you think about prokaryotic cell context it is a single DNA molecule.

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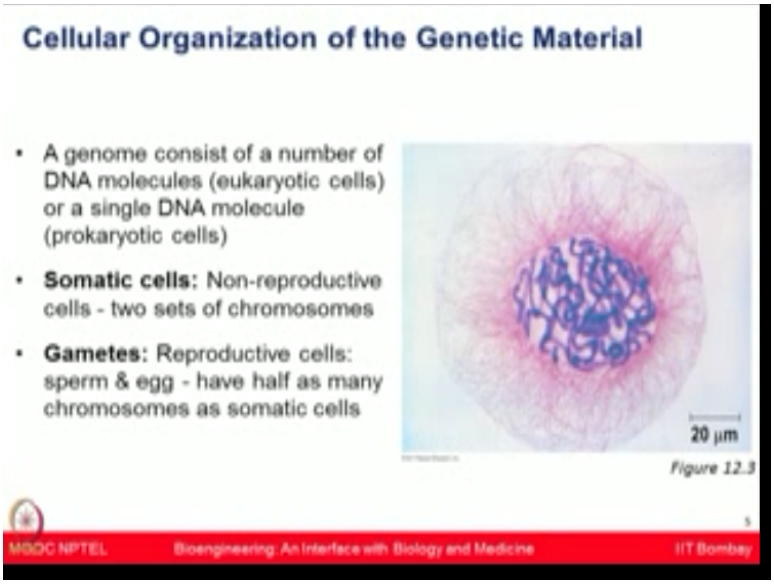

Now let us also get some definition for somatic cell. These are the normal body cells non reproductive cells, which contains 2 set of chromosomes whereas the gametes they are the reproductive cells which are especially sperms and X and they contain half as many chromosomes as the somatic cells. Let us not talk about cell cycle.

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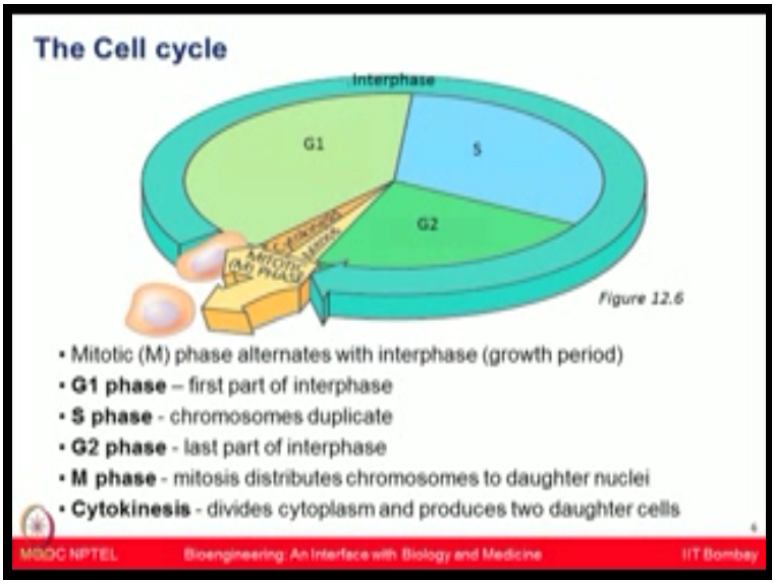

In the cell cycle, the mitotic phase alternates with a large growth phase which is interphase. Now this entire cell cycle could be divided into multiple phases. Let me go to board and draw you the cell cycle.

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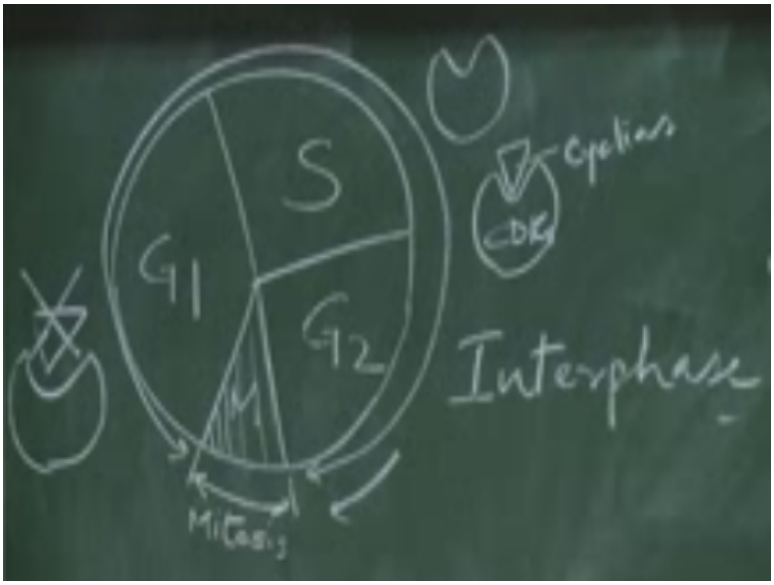

The cell cycle 1 of the phases of the cell cycle is known as G1 phase and then we have S phase and G2 phase and then we have a short mitosis phase. The mitosis phase is much smaller, but rest of the part which is essentially the interface or the growth phase that is much bigger phase, right. So the entire mitosis process which is going to be responsible for cell division to happen that is actually much shorter phase.

But remaining phase which you can see that is the known as interface that is much larger phase. So this is interface which prepared the cell and ensured that the cell is ready for division to happen. So the mitotic phase alternates with much longer interface and it is almost 90% of the overall cell cycle, which comes in the part of the interface. Now the G phase link actually comes little misnamed, especially the gaps but these are not the gap these are actually quite crucial for cell to prepare.

And G1 phase ensured that all the, you know, the nucleic acid content the chromatins are sufficient, present and then now cell is ready for DNA replication to happen in the S phase and then the G2 phase is there which now ensures that the DNA replication has occurred properly and cell is ready for further division to happen in the mitotic phase. So now the same concept you can see in the slide that we have G1, S and G2 phase.

The G1 phase is first part of the interphase, S phase is where the chromosomes get duplicated, the G2 phase is last part of the interface and M phase is where the mitotic division happens and this process distributes the chromosomes to the daughter nuclei. In the process of cytokinesis then the cytoplasm gets divided from the 2 daughter cells and now the 2 cells contain you know the same cytoplasm and now they have a bifurcation from the parent cell. So let us look at broadly the mitosis and meiosis processes.

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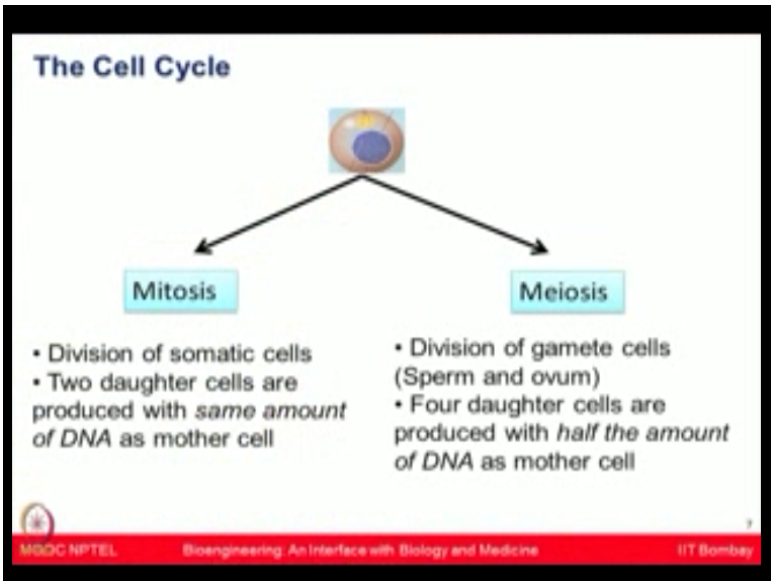

In case of mitosis, the division of somatic cells happen whereas the meiosis governs the division of gametes especially the sperm and the ova. In mitosis the 2 daughter cells which are produced they contain the same amount of DNA as the mother cell. In case of meiosis 4 daughter cells are produced which contains half the amount of DNA as the mother cell. Let us first start discussing about mitosis.

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And why mitosis is so important. So think about all our you know the growth requirement many times if you know the old cells get replaced. The old cells get replenished. The damaged cell has to be repaired and you know new cells has to be produced. All this only happens because of the process of mitosis which is the process in which the cell divides to form 2 identical daughter

cells. Let us first talk about few basic terminologies before we actually talk about the mitosis process.

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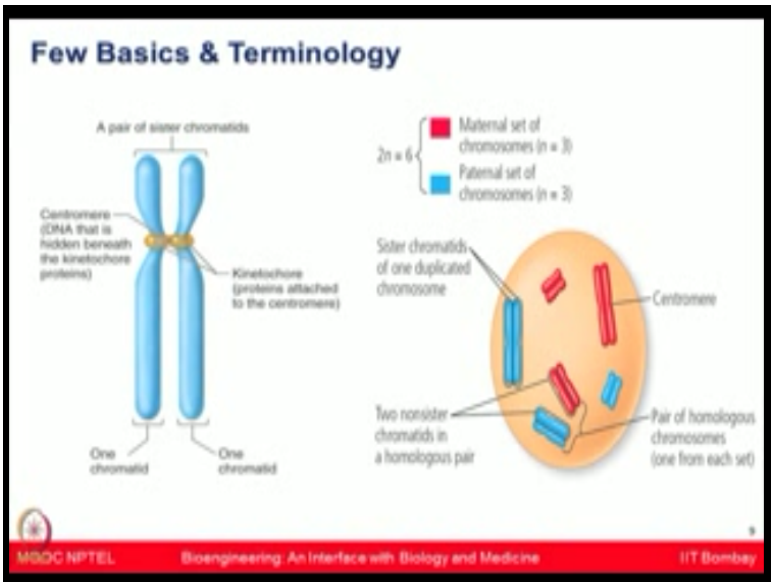

So for example in the blue color on the left side, you can see a pair of sister chromatids are there and then we have this DNA, which is hidden behind this you know the protein complexes. That region is known as centromere and the protein which is surrounding to that is kinetochore, which is the protein attached to the centromere region and this chromosome has the pair of sister chromatids. So you know the 2 chromatids are shown in the blue colors.

Now let us think about you know a cell which is having 3 chromosomes each from father and mother. Let us say the maternal set or the mother obtained chromosomes are shown in the red colour and the blue one is shown from the father. So the 2n or the ploidy level is 6 chromosomes in this case. So each chromosome is having these sister chromatids, which whether it is the blue ones or the red ones which are connected with the centromere.

And now when they form the homologous pair that is you know the two, the both the blue ones and the red chromosomes comes together that is the chromatids from the non sister chromatids of the homologous pairs are being formed all you can say that you know these are pair of homologous chromosomes from both one from each of the paternal and maternal sets is shown in the circle here.

You know another interesting fact is that how DNA is so, you know, so much DNA content is present, but how it is so tightly packed in the nucleus of every cell in a such a short space. So it is not only DNA, but they know in addition to DNA there are separate histone proteins, which are also attached to the DNA molecules and these you know 8 histone proteins along with the DNA molecules they form a complex which is nucleosomes.

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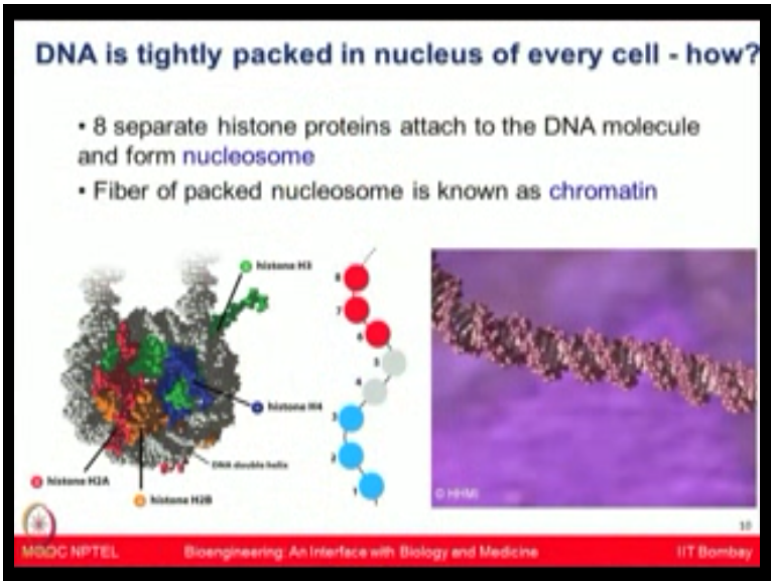

And these you know the fiber of these packed nucleosomes that is known as chromatins and these chromatin they get condensed before the process of mitosis starts. So in the image shown here, is various histone protein like histone h1, we have histone h2a, h2b, h3 and h4 and you can see that how 8 histone proteins are you know packed up and informing the nucleosome and meaning of these nucleosomes together will form the overall chromatins. So let us have the broad overview of mitosis process first.

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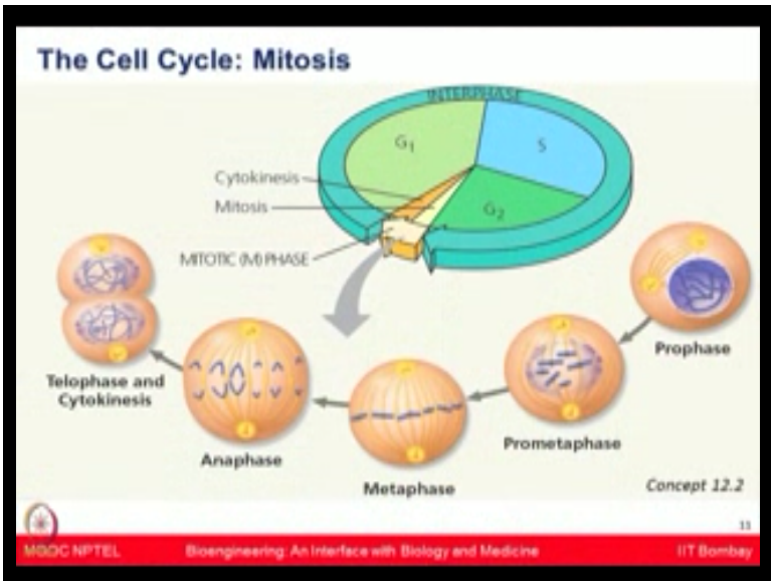

So thinking about you know again the overall cell cycle, we have this my mitosis phase and we have the interface. Now within mitotic phase, then there are different phases involved for example prophase, prometaphase, metaphase, anaphase, telophase and cytokinesis. Let us go each one of them in slightly more detail one by one. So first let us look at you know thinking about this nuclear content, they are getting condensed and then they are forming the chromatins, right. So that is what is shown in the blue color here.

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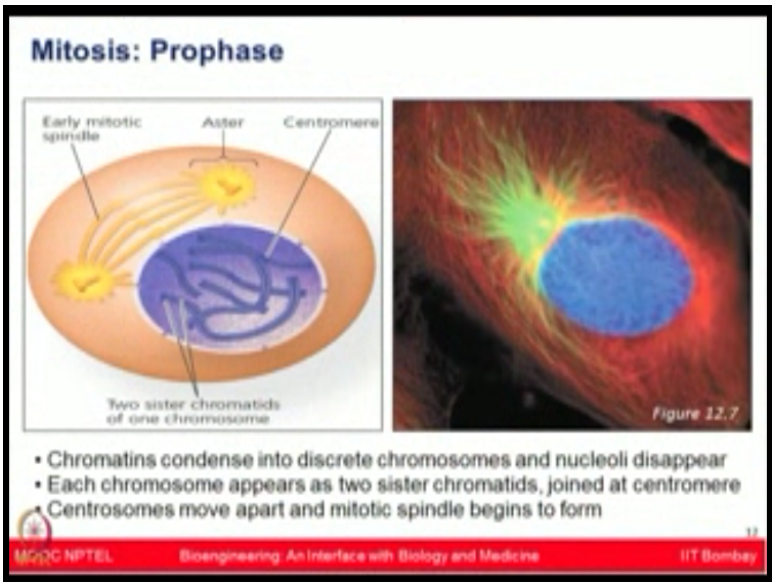

The sister chromatids are being formed and converting the condensed material is forming other discrete chromosomes and during this process the nuclear get disappeared from the cell. So each

chromosome appears as 2 sister chromatids and they are actually joined at the centromere. Now then at the same time the central zones, they move apart and the mitotic spindle starts to form.

Now you can see this particular electron microscopy image, which kind of you know shows that how people make observations in the actual research looking at the cell images and that is what is then drawn in the cartoon form. Now let us move on to the next phase which this prometaphase. Now cell is trying to prepare itself for division to happen and start orienting it toward the 2 poles and that is the time when our nuclear membrane starts fragmenting and the microtubules they starts growing.

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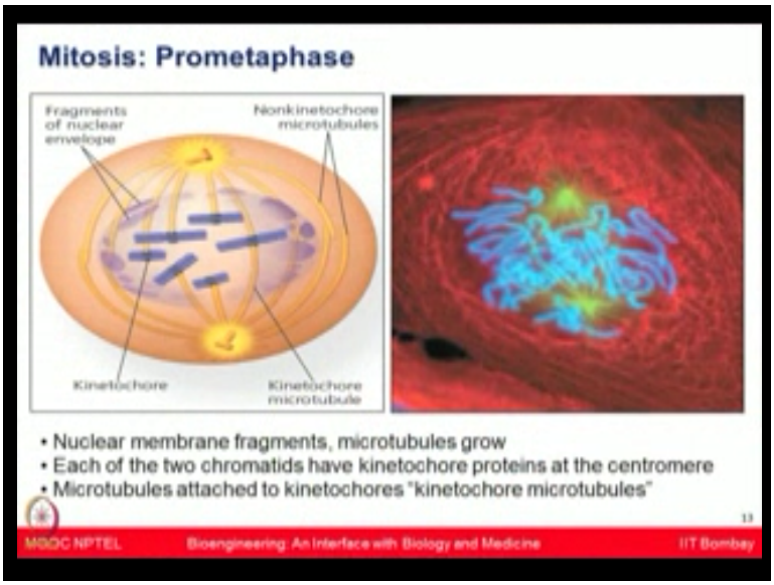

Then each of the 2 chromatids which have the kinetochore proteins at the centromere. Now this microtubules get attached to those kinetochores and they are known as the kinetochore microtubules. So again there are lot of shuffling is happening in the cell. All these you know the chromosomes and these chromatids from these chromosomes they start trying to align themselves towards the metaphase plate, which is just in the center, but they are still not aligned.

But they are trying to align themselves, but then the cell moves to the next phase, which is metaphase.

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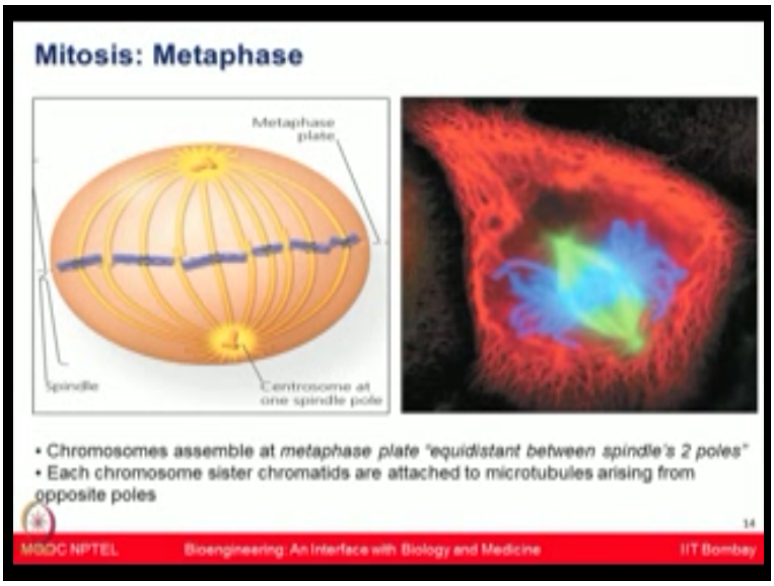

Now at this time the chromosomes they get assembled at the metaphase plate which is equidistance between the 2 spindle poles. If you think about from 2 spindle poles from the top, then just in the center that equidistant region is where the metaphase plate is formed. So each chromosome sister chromatids they are actually attached to the microtubules which are arising from opposite poles.

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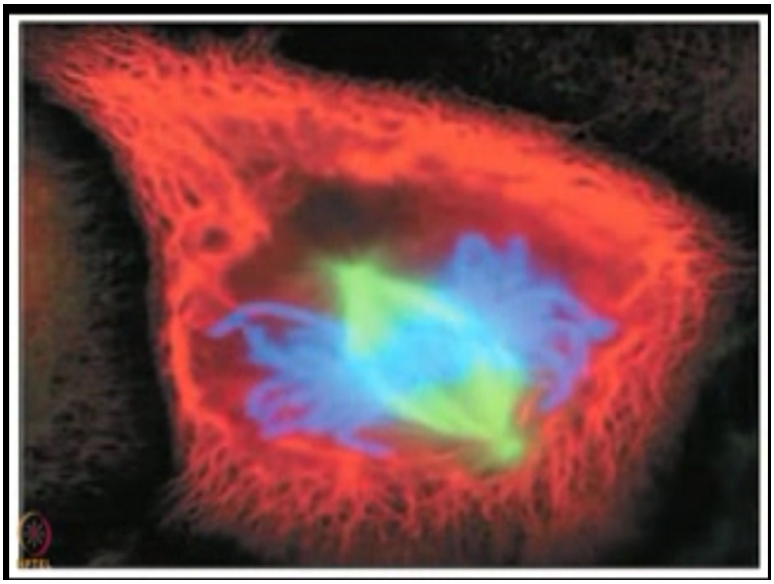

Now you can see the same in the microscopic image alright. So now let us move on to an anaphase. During the anaphase, the sister chromatids they separate and now each chromatid, they behave as a chromosome. So during this process things are not so straightforward. You know a set of protein the complex proteins are involved, especially the coalescence they help in the

process of cleavage and then the daughter chromosomes they move towards the opposite poles. Now similar concept now can be seen in the microscopic image on the screen.

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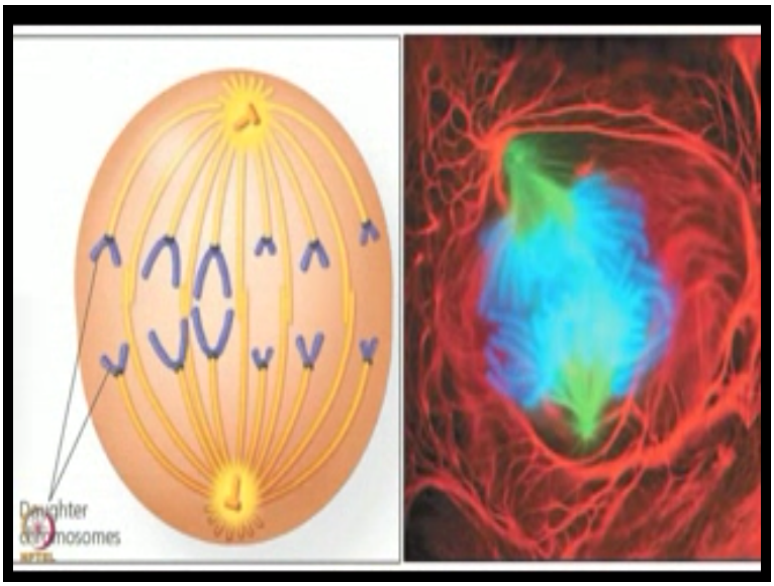

So then the last part comes which is the telophase and then the cytokinesis happens. So for example 2 daughter nuclei form in the cell. Now the nuclear envelope start reappearing because they want to now produce 2 daughter cells. The spindle microtubules they get depolymerized and then the chromosomes becomes less condensed at this time and at the same time now that karyokinesis process or division of nucleus also gets completed.

So the microscopic image shows you that now you know from 1 cell now the 2 cells started you know appearing, although distinct boundaries are still not made and that happens in the process of cytokinesis.

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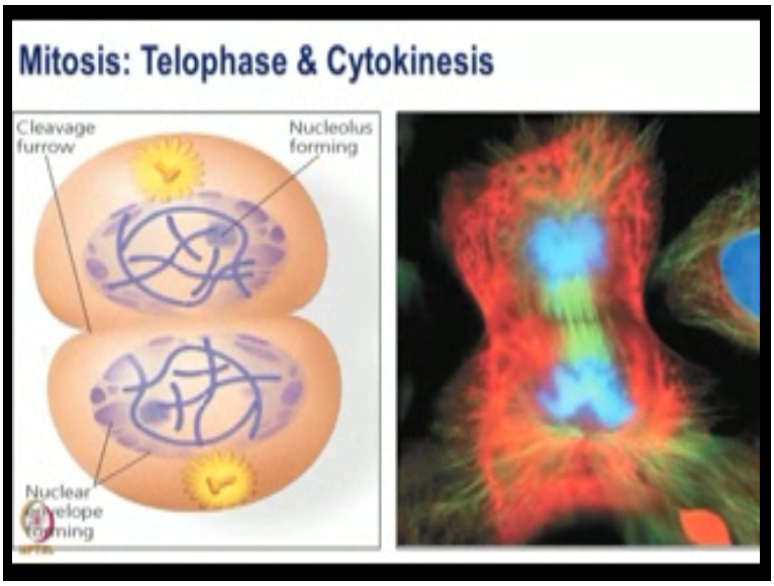

So at the process of cytokinesis, the cell furrow is being formed and now the division of cytoplasm gives rise to 2 daughter cells. So now let us kind of you know review the whole thing again. We talked about prophase, we talked about metaphase, then metaphase, anaphase, telophase and cytokinesis. So this completely you know how from 1 cell, the 2 daughter cells are being produced having the same nuclear contents and that kind of you know showed that your body is growing.

There is a sufficient development is happening. Now let us move on to another important process which is meiosis.

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So what is meiosis and why meiosis is important. So think about you know if the cells are dividing and you know in the case of meiosis if the numbers of the chromosomes are getting reduced half and if the process is so mechanical, then every offspring from the same parents will look exactly same right. So if now, let us imagine that you know there are four children from in one family, so from the same father mother then they will look exactly same.

But there is actually a lot of genetic variation. So question is from where the genetic variability comes. So the ability to generate these kind of genetic variations in the children because the process of meiosis happens randomly and at that time lot of gene shuffling happens across the chromosomes and that actually you know ensure that each one of us is very unique in our appearance, in our identity, in our entire genetic components. So meiosis is very central to the reproduction process.

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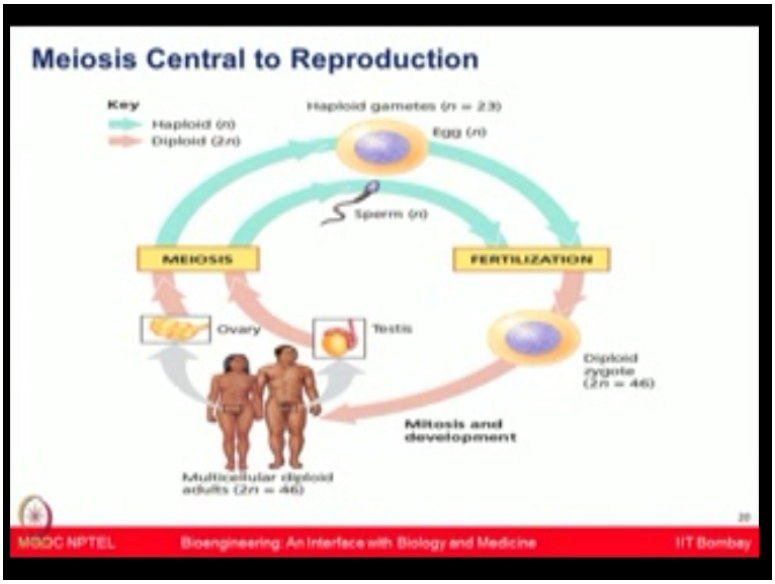

And if you look at the cell here, we have starting from this firm and the ova, which is produced inside the testes the sperm and inside the ovary these X are being produced. Now these are haploid shown in the green color. After fertilization process, they form the diploid zygote, which is now the 2n. So what we can see, we can see the ploidy level here that you know one phase is having haploid level.

And now the other part when after the fertilization and the mitosis and development which happens that is a diploid level. Now the multi cellular diploid eggs are being formed which are having 2n equals to 46 chromosomes. So let us now move on to meiosis, but considering the time constrain and thinking about you know what is so crucial for you to understand, let us not talk the entire meiosis process in detail, but rather try to compare and see the what are the salient features which are different in meiosis as compared to mitosis.

So mitosis we have talked in some detail and you have some good idea now. We will now compare that what are the key differences of mitosis and meiosis.

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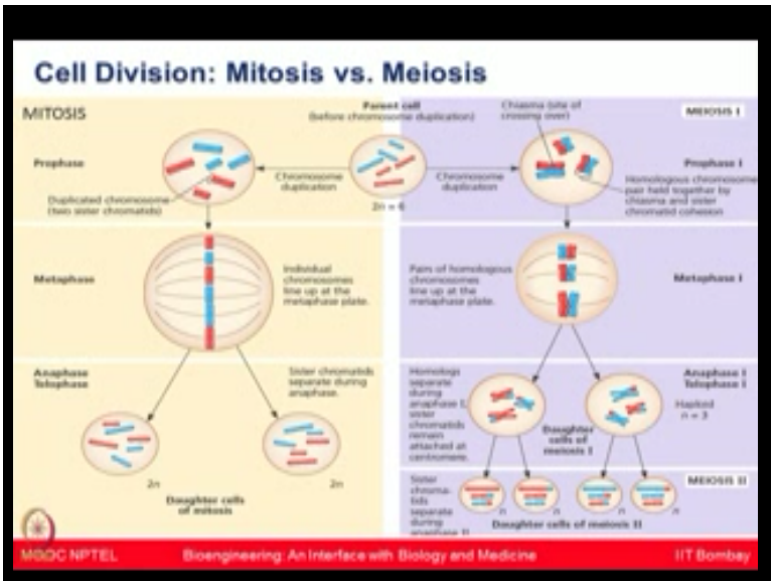

So for example in meiosis as opposed to mitosis, there are 2 divisions happens meiotic 1 and meiotic 2. Now meiosis 1 having certain peculiarity but meiosis 2 is exactly like the mitosis. So now let us take the same parent cell which we know took in the beginning for the definitions purpose, which has a 3 red and 3 of the blue chromosome from maternal and paternal sets. Now let us review again the mitosis process. For example, the chromosome duplication happen.

Now you can see these you know the condensed these chromatids on the left side in the prophase, 3 blue ones and 3 red ones. Now these duplicated chromosomes, they are forming 2 sister chromatids, then after the prometaphase and the metaphase, now these individual

chromosomes get lined up at the metaphase plate, which is equidistance from the spindle poles and now in the process of anaphase and telophase, the division happens.

And now 2 daughter cells are produced which are having 2n exactly the same like sex chromosomes where you started right. That is what the left side of the image conveys which refreshes your understanding for mitosis. Now let us try to compare a few things in the meiosis. So in meiosis now as you can see the blue and red chromosomes, they are you know having some sort of interaction and in certain region there the crossing over happens.

So this crossing over is known as a chiasma, which is the site of crossing over and that is the time when you know some of the genes start getting shuffled from the blue to the red ones, so from the maternal and paternal side. Now we have few genes which are going to shuffle to both the chromosomes and now these homologous chromosomes they are actually you know pairing and there is still held together by the chiasma and now the sister chromatids by the coalescent proteins.

So this entire thing is slightly different as compared to what you see in the prophase of the mitosis. So let me highlight you know the 3 distinguishing feature here, which makes meiosis more unique. The very first one revolves the crossing over or the formation of chiasma. Now these homologous pair of the chromosomes, they are itself moving and they are forming the metaphase plate when they are actually you know getting lined up at the metaphase plate.

So in this case now not the individual chromosome, but rather the pair of the homologous chromosome they are reaching to the metaphase plate metaphase 1 and then now these homologs they get separated when the anaphase 1 and the telophase 1 processes are happening and therefore now the daughter cells, which are produced they are having the you know both red and blue patterns together right.

And then further once you have finished your meiosis 1, then the entire processes of mitosis getting replicated and on meiosis 2 you are going to produce 4 cells out of 2 cells. So then you have daughter cells of meiosis 2 which are 4 cells now and n become becomes half. So you have

only three chromosomes, but each of the three chromosome as you can see they do not have very clear red or very clear blue, you have the patches of fluid red and blue.

Which means the genes have shuffled and the genetic variability has been introduced. So to just summarize the first part of the lecture, we have first you know tried to understand the cell cycle broadly, the very small part of mitotic phase, which is so crucial and how the cell divide to form the 2 identical daughter cells. We have also tried to understand that how meiosis generates the genetic variability in the off-springs.

And now I think we are going to continue our understanding about the cell cycle, but more in the context of its regulation, dysregulation and how the disease like cancer may happen. So now let us talk about the cell cycle regulation, its deregulation and how disease like cancer may happen.

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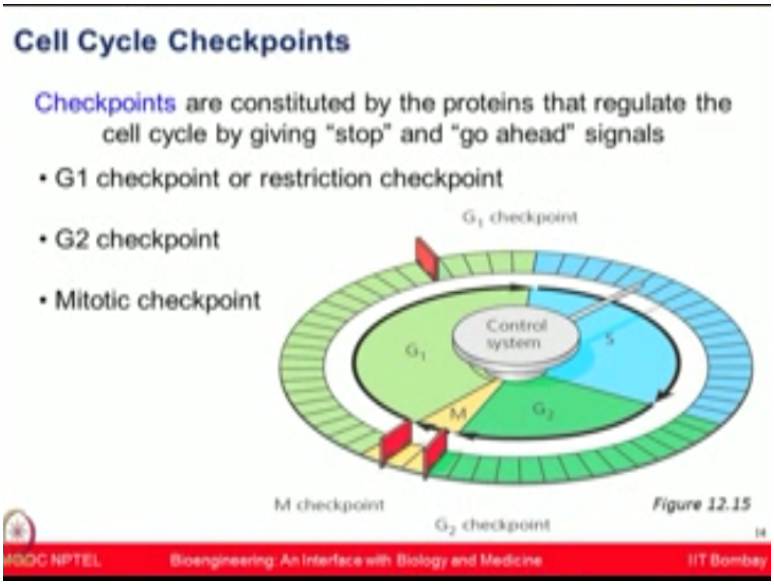

First let us look at the cell cycle checkpoints. Just imagine that you know whether cell cycle is being operated. You want certain you know checkpoints to ensure that the entire cell cycle is governed properly. Just imagine that you know when you are driving on a road, you need those kind of you know the speed breakers, you need the traffic police to ensure that everybody is obeying those rules.

Otherwise you know sometimes things may go unattended and accidents may happen right. So like that, there are certain checkpoints even in the cell and there is no police inside their cell. So there are certain you know proteins which are following the same pattern of what you know to regulate these particular signals. So the cell cycles are actually having the checkpoints, which are constituted by the proteins which regulate cell cycle by giving either a stop or go ahead signals.

Then there are different you know checkpoints involved.

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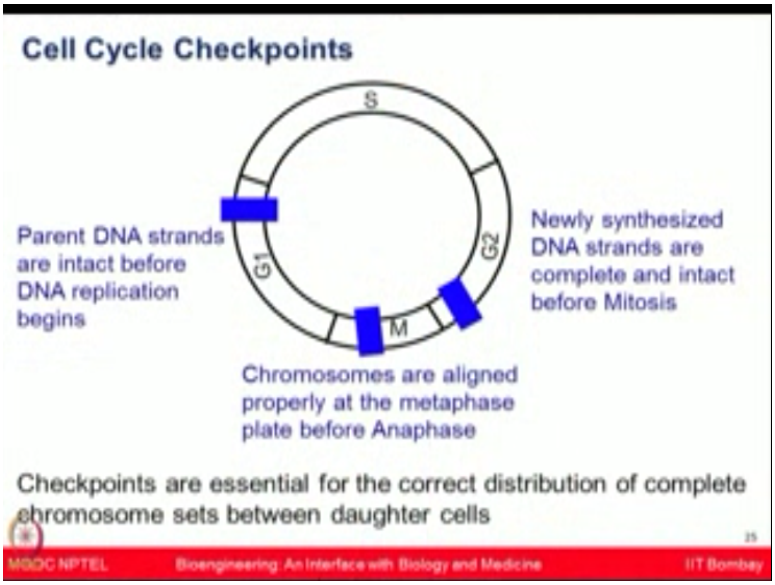

For example, even the G1 phase there is a checkpoint or recession point, which ensures that now the, you know, the chromatin material is sufficient, the cells are getting condensed with those nuclear contents and it is getting ready to move to the S phase for DNA replication to happen. Then there is a G2 checkpoint, which now ensured that now cell is totally ready for the mitotic division to happen and then a mitotic checkpoint is there or the M checkpoint is there, which ensures that the mitotic division has happened properly.

So as you can see there are different checkpoints, which are involved to ensure this process is happening sufficiently. Now how these things happen, so as I already mentioned the G1 phase the parental DNA strands are intact before DNA replication begins. So the G1 phase checkpoint ensures that this process is done properly, then the G2 checkpoint it ensures that newly synthesized DNA strands, they are complete and intact before the mitotic process to happen.

And then in the metaphase plate, the M phase checkpoint the chromosomes are aligned properly at the metaphase plate before the anaphase happens. So this is kind of you know various checkpoints, which are so essential for the correct distribution of complete chromosomes set between 2 daughter cells. Now let us think about you know this entire intricate process, which is so complicated how it is being governed by certain proteins right.

So a set of proteins known as cyclins and a cyclin-dependent kinases they play an important role to govern these processes.

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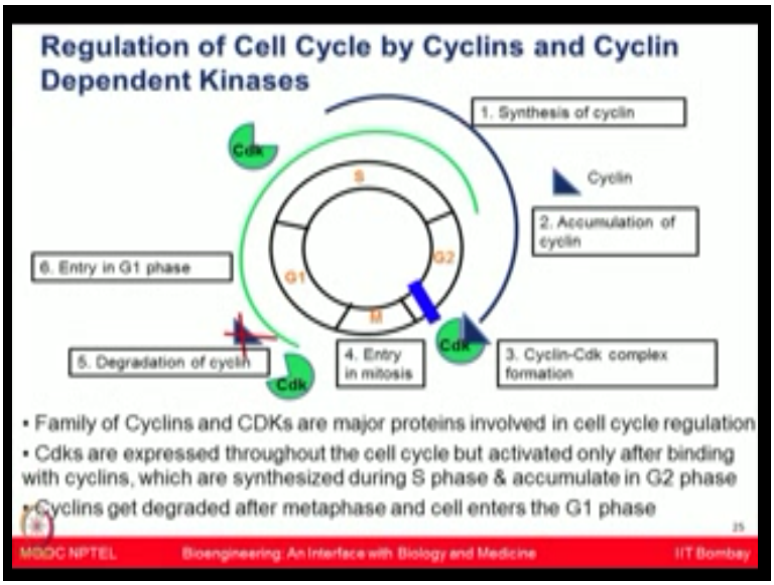

Let us now go back and in our cell cycle diagram, now let us draw that you know how these proteins you know where they are being synthesized, all right. So the cyclin proteins are synthesized in the S phase right and now they are getting accumulated, as they are moving into the G2 phase and then at that time they are interacting with CDKs or cyclin dependent kinases to form a complex which is CDK and cyclin complex.

Now this ensures now that inner cell is now ready to move for entering the mitotic process. Now once the mitosis is finished, then the cycles get degraded and now you know this complex get dissociated. Now the CDK got separate and cycles got degraded. So this ensured that now the

mitotic process has finished. So in this way just by using you know these proteins, cell is ensuring that this particular cell cycle is governed properly.

So let us look at this in this diagram. Now the synthesis of cyclins happens in the S phase. Now the cycles are getting accumulated and then the cyclists are forming a complex with CDKs, then they are allowing the cell to enter in the mitotic process and then the degradation of cyclins happen and CDKs get dissociated, which ensures now this cell has completed that particular part. So this now moves the cell into the G1 phase.

So these are the family of cyclins and CDKs which are you know very crucial for cell cycle regulation and these CDKs or cyclin dependent kinases, they are expressed throughout the cell cycle; however, they are activated only after when they are bound to cycles, which could be synthesized during the S phase and gets accumulated in the G2 phase. As I mentioned that cycles get degraded after metaphase and now the cell enters in the G1 phase.

So in addition to these, there already know various external signals which also regulate cell cycle. So what could be you know some of the internal signals and external signals. I think let us discuss about them.

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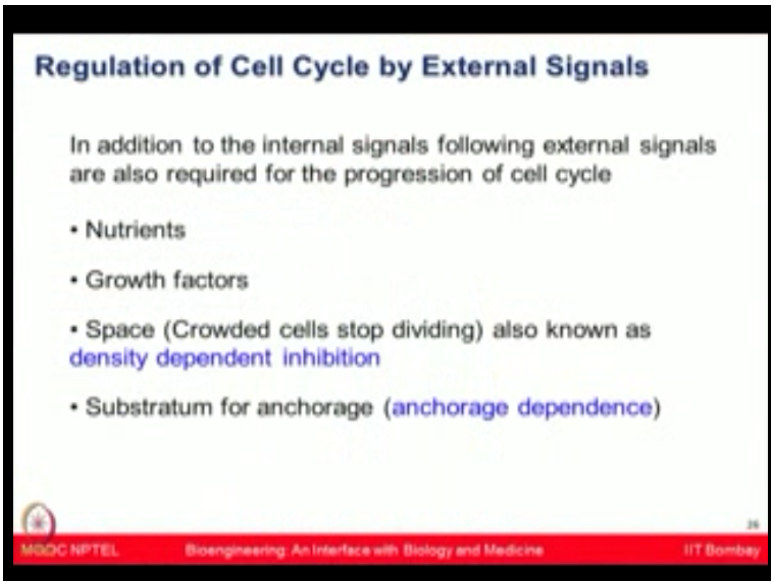

So for example nutrients, growth factors, even in space and the substrates, all of these things are also ensuring in which cell cycle is regulated. So just imagine that you know nutrients and growth factors are of course you know crucial for the cell cycle to operate, but even otherwise you know even the space can be constrained like you know how much space is available. If it is too crowded and there is not much enough space, then cell should ideally stop dividing.

And that is the process known as density dependent inhibition, because there is not enough space now for more cells to grow and that is where you know the difference comes to a controlled growth versus uncontrolled growth. In case of normal cells now, a normal will stop growing when they see these kind of density have been achieved. Then density dependent inhibition is happening whereas in case of cancer cell they are not going to get affected.

They are not going to obey this rule and they are keep continuing to grow and they are then going to form the tumor mass. Now additionally, these cells are going to attach to certain environment, certain substrates for their anchoring and that is known as anchorage dependence, you know way for growing the cells. So again this part is not being obeyed by the cells in case of the cancer cells.

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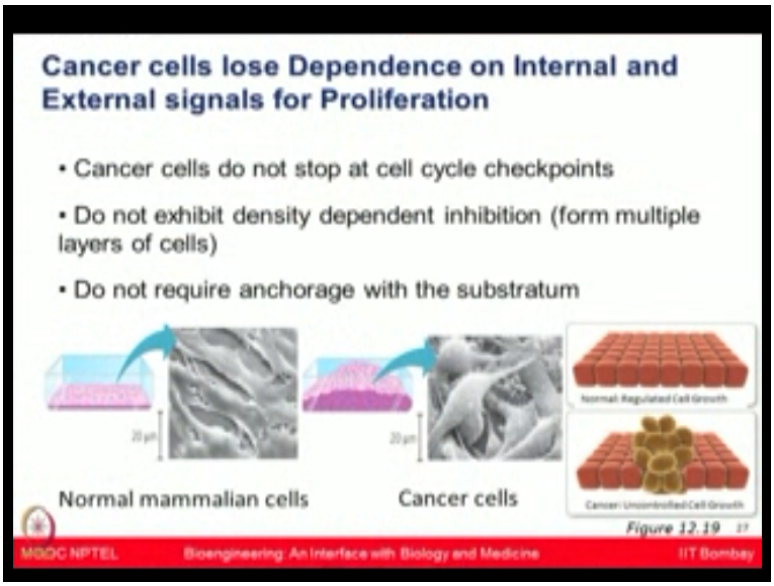

So now let us look at what happens in case of the cancer cell, which they lose dependence on the intern and external signals for the proliferation. So cancer cell, they do not obey the normal rules

for the cell cycle. They will not stop at the cell cycle checkpoints. They will not exhibit any kind of density-dependent inhibition and therefore they will form the lump, the tumor masks with a multiple layer of the cells.

They do not require any anchorage with a specific substrates to get attached and you know therefore they can actually grow much happily on any environment and now they can you know also they are uncontrolled in growth. They are not obeying the cell cycle checkpoints. So therefore they are actually you know in some way, they are not at all following the normal cell cycle rules.

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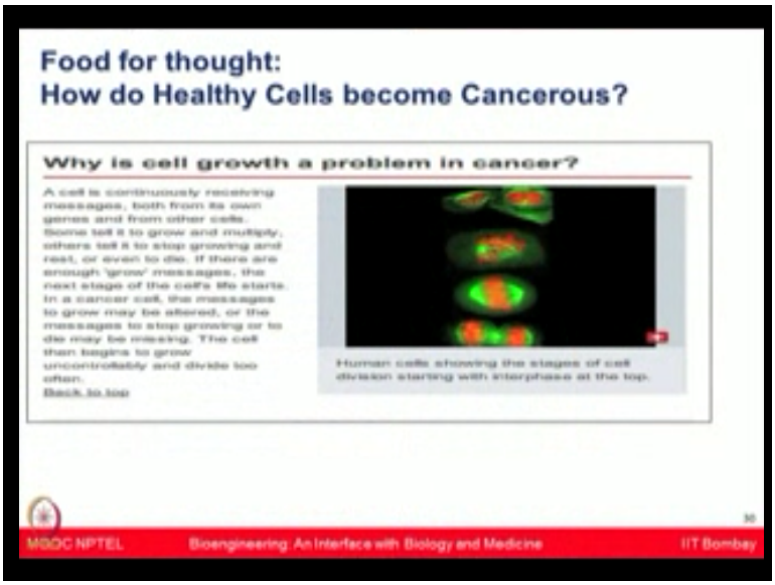

So there are couple of food for thoughts from a journal, which shows here that you know how do healthy cells become cancerous and these are still an area for research. We have to still think about why the cell growth is problem in cancer, how long do cancer cells live for, why are the cancer cells so powerful.

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So I do not have time to talk about each 1 of this point. I will give you reference. I think you should read this paper and you should try to understand that you know how much challenge we have is still in science and to do research in this area. We have still not understood these cell properties so much. Why do not cancer cells die normally, why we cannot control them, what are the missing checkpoints in these cancers and how do cancer cells they escape destruction.

So all of these are the food for thought for you and I think you have some reference to read, but there is still too much research to be done in this area. Now what is important again to emphasize that there is so much need for you to know the basic biology, but also implement that at a technology level that we need detection technology, the clear detection technologies to detect the cancer cells early.

So there is a lot of emphasis on looking at even circulating tumor cells with the new detection technologies and in United States Food and Drug Administration, in the USFDA, they have cleared some technologies, for which allows you know very sensitive capture of circulating tumor cells.

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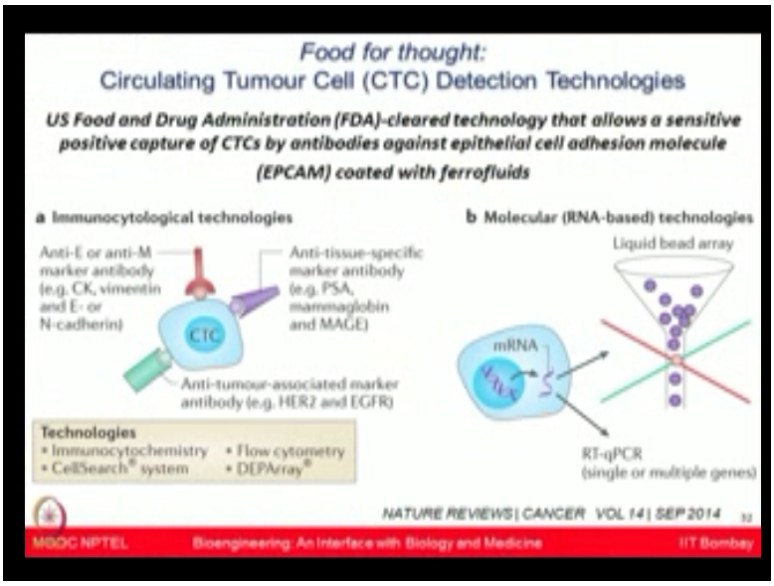

Which is looking at you know certain antibodies against the EPCAM or epithelial cell addition molecules which are coated with certain ferrofluids. I am showing you again from a review article 2 different approaches here. One is immunocytological technologies and second is a molecule based technologies, in which way by looking at certain markers some of the proteins, which are found in these tumor cells and an antibody generated against them.

How this understanding, this knowledge could be utilized to build the devices, which could detect these circulating tumor cells ahead of time and much more accurately. So these kind of you know technologies will be required. I am again going to give you a reference to read this paper. I am not talking in detail right now.

But just illustrate that basic understanding, basic science is revealing the right targets, the right proteins against which antibodies could be generated and then the technologists have to come into the play to find out a way to develop the devices, the detection system which could accurately detect these cells and is there a way now.

Just imagine the challenge for you, can we now you know if out of you know let us say 1000 cells 1 of the cell is showing you that the tumor behaviour can you, you know, with any kind of magnetic property can you exclude that out from the normal cells. So then can we now control

the cancer if we are having this kind of technology which can detect the tumor cells, which can also exclude them out of the normal cell circulation.

These are the challenges for us, but I think an area which needs lot of exciting research. So in general today in conclusions, we have discussed about broadly about a cell cycle and you know various you know mitotic and mitotic phase.

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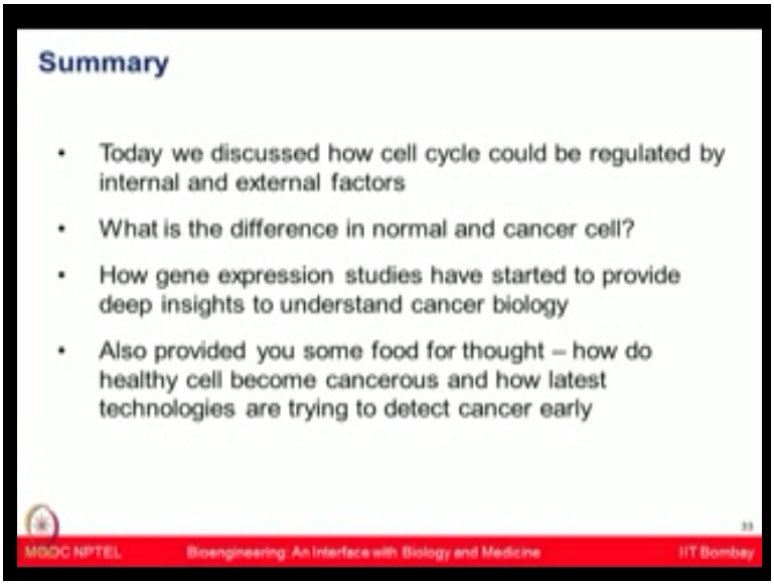

There is a kind of contrasting features of mitosis and meiosis different internal, external factors which govern the cell cycle. I am just trying to kind of convey you there is some basic differences between normal and the cancer cells and I am sure you know you appreciate that there is lot of fundamental research going on to look into various type of you know the gene mRNA and protein level changes in normal and cancer cells to find out the right marker proteins.

Which could you know lead us to find out the good detection technology to detect the tumor cells and I have shown you a couple of you know food-for-thought, some challenging questions in the area of cancer biology and also some of the promising technologies, which are coming forward which could be very helpful in the translational research in future. Thank you very much.

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References

- Campbell Biology - Reece, Urry, Cain, Wasserman, Minorsky, Jackson 10th Edition, Pearson

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