Genetics & molecular biology

◉ Sheet ○ Slide

Number:	-35
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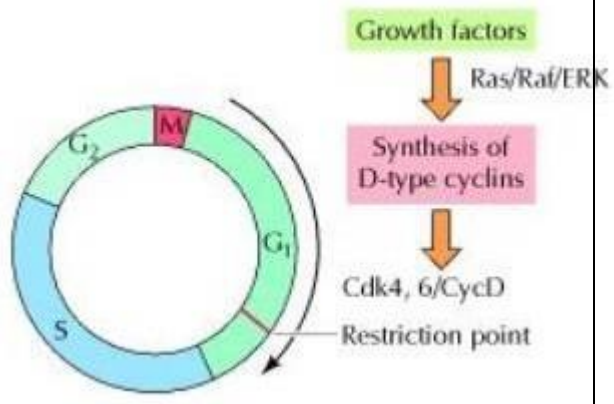
In this lecture the doctor talked about the cell division and apoptosis mechanism



-The cell cycle is divided into 3 phases G1, S, G2 then mitosis which consists of: Prophase Metaphase Anaphase Telophase and the Cytokinesis.

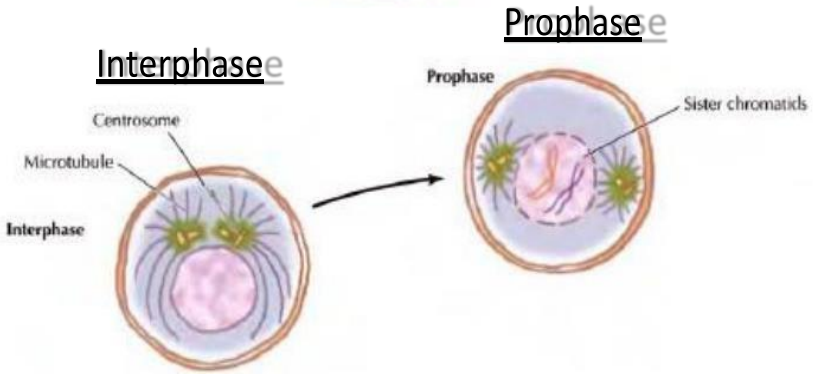
We'll discuss each one,

When the cycle is done with the G2 phase it will enter the prophase, the chromosomes are still loose in structure they are not separated and you can't observe

each one alone so first thing happens is the condensation of the chromosomes and each

chromatid is connected with its sister chromatid with a centromere

here we can detect the X shape of the chromosomes.

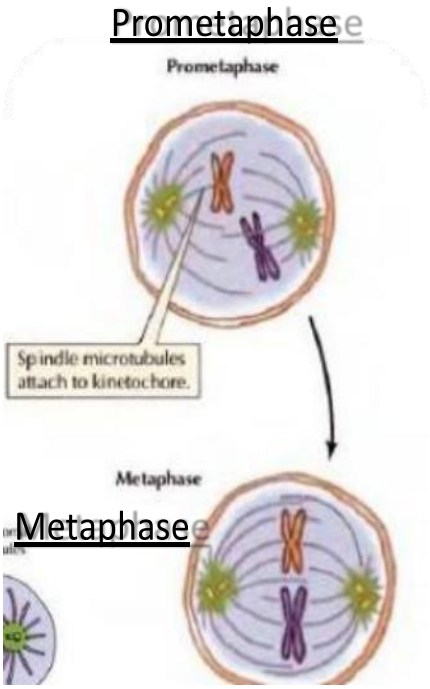


What's the purpose of condensation? It helps with the segregation of these chromosomes

so its like a quality control to guarantee that each sister chromatid is divided completely into each daughter cell

The centromeres are DNA structure that contain a non-coding region(theres no genes) this region will bind to protiens to form the kinetochore and in the kinetochore the attraction of the spindle fibers will take place the centromerewill undergo duplication (like any other sequence in the DNA during the s-phase of the interphase so that when they divide both daughter cells will have a centromere and they will move to the opposite sides one copy will move with its sister chromatid to one pole of the cell and the other will move to the opposite pole of the cell and to specify which pole of the cell the sister chromatids are going to move the spindle pole bodies will embed right under the nuclear envelope.

1-Prometaphase and metaphase:



Now that the chromosomes are condensed they will start to align

(this is the shift from prophase to metaphase) in the middle of the cell to form the

metaphyseal plate they are arranged above each other.

When they are completely aligned we are in the metaphase

They are going to keep shuffling back and forth until they reach

the middle and they are aligned and here is another advantage of condensation,

we are limited by the space we can only align the chromosomes in the middle and

not anywhere else in the cell so we need to condense them to decrease their size and occupy a little space.

The kinetochores will attach to the centromeres and

they will start to move the chromosomes on the spindle fibres

the chromosome is moving (such as the movement of the vesicle),its not the depolarization of the spindle fibre that pulls the chromosomes, it's a movement by the motor molecule carrying the chromosomes toward the poles of the cell.

These spindle fibres that are made from microtubules they are oriented in the

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Note:

-The kinetochore is the site of spindle attachment

-The metaphase plate is the lining up of

chromosomes single file in the center of the cell

2-Anaphase

In this phase the movement starts and the separation of the sister chromatids will take place to the two opposite poles of the cell.

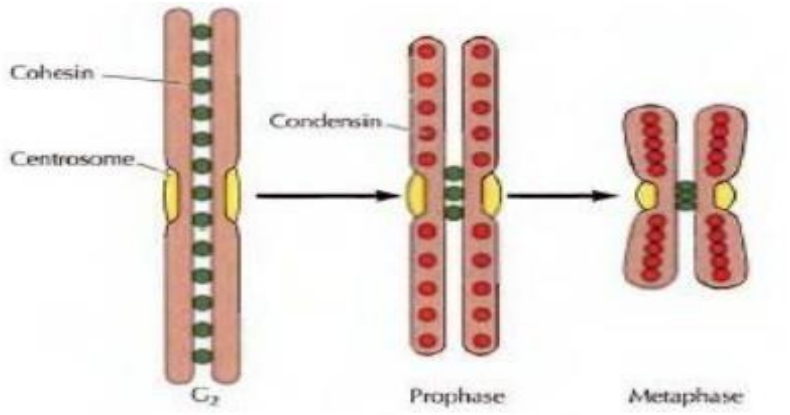
3-Telophase and Cytokinesis

Reformation of the nuclear envelope that wraps the genetic material and constriction in the middle of the membrane to form the contractile ring that's made of actin filaments and myosin then complete separation of the cells by cytokinesis.

In the book they convent that the Mitosis is only the separation of the genetic material and formation of the nuclear envelope, but the separation of the cells and formation of the contractile ring is the Cytokinesis.

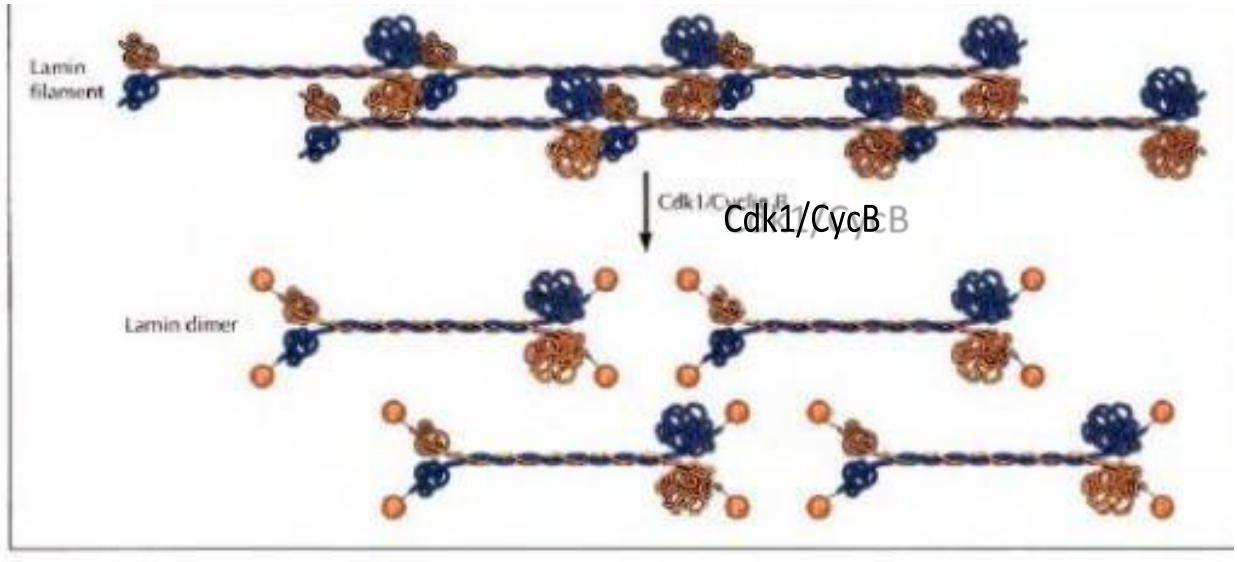
1-When the cell finishes the cell cycle and its in the G2 phase, replication is done so as a quality control we need to hold the sister chromatids together, so that when its time for mitosis the sister chromatids are already held together, so in the G2 the sister chromatids are held together by a protein called cohesin and its holding the chromosomes from the beginning till its end, all the surface area between the two chromosomes is held by the proteins.

2-When the cell is in the cell division phase the connection between the two sister chromatids will change to another protein called Condensin and the location changes too,it only hold the two chromatids between their centromeres. It helps in keeping the connection and facilitate the separation because the surface area of connection is less. One more important reason is that if the cohesin is still found between chromatids that chromatids cannot compact or reduce their sizes for the same reasons I mentioned above (metaphesial plate)



Breakdown of nuclear envelope:

The nuclear itself is held as a structure by the action of the nuclear lamina, disassembly of the nuclear lamina takes place by phosphorylation, by the action of cyclin independent kinase (CDK1) that associates with cyclin B. CDK1-Cyclin B complex is formed in the G2 phase.



Depolymerization of the nuclear lamina takes place and fragmentation of nuclear membrane and dissociation of the pore complex.

Apoptosis: Programmed Cell Death

Is the exact opposite of cell division, its planned, organized and normal physiological process not necessarily a cell that dies with apoptosis to be abnormal.

Uses of Apoptosis:

-During development some cells dies by apoptosis and some cells persists until a structure or an organ develops.

-Regeneration in adults in general uses apoptosis. Regeneration of RBCs and intestinal cells that are fully mature does not divide(in G0 phase),and if I want new cells I use stem cells, so eventually if the body wants to get rid of these fully mature cells, apoptosis is the solution.

-Getting rid of faulty synapse if the neuron is misplaced by getting rid of neurons

Microscopic features of apoptosis:

1-DNA fragmentation and chromatin condensation, we see a dark nucleus

2-Fragmentation of the nucleus (it looks like we have more than one nucleus, so we have to make sure if we're examining a multinucleated cell or a regular apoptotic cell.

3-Fragamnetation of the whole cell

**The cell that went through apoptosis will be detected by microphages as cell debris, so the cell must have a tag, this tag is the flipped of phosphatidyl serine, normally they are found in higher conc. in the inner leaflet so they'll be flipped and recognized by receptors of macrophages and phagocytosis takes place

Activation of Apoptosis inside the cell;

There is more than one pathway that eventually leads to apoptosis.

A family of protiens named BCL-2 will be activated, some of them are inducers and some are inhibitors(BCL-2) of apoptosis.

When activation takes place a pore structure within the mitochondrial membrane will be made and allowing the exit of cytochrome c,it will bind to adapter protein which then caspase-9 will bind to the adapter protein, activation of caspase 9 will take place.

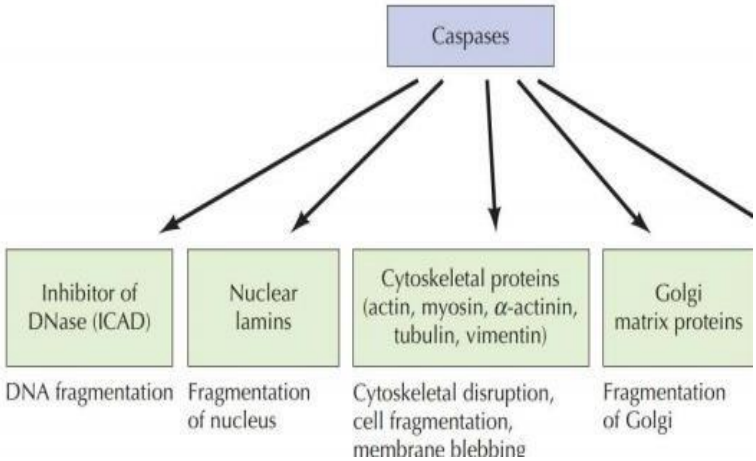
Caspase 9 is a protease which will start to digest and cut cellular materials auto cleavage will occur and its activation will activate more caspases (caspase-3).

(caspase-3 in found as procaspase 3 and its cleaved into caspase 3 by the activity of caspase 9)

The caspases have the same concept as lysosomal enzymes they are secreted inactive, and activation takes place by cutting a part of them

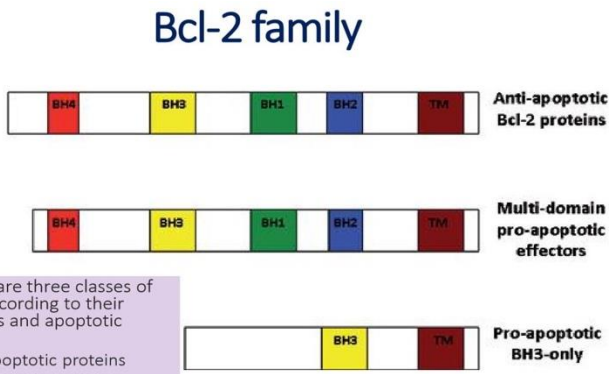
Caspases roles:

1) Disassembly of cytoskeleton(actin,intermediate filaments) to allow fragmentation of membranes
2) Degrade the nuclear lamina to degrade the nucleus
3) Inducing dna fragmentation by activating DNAase by inhibiting the inhibitor of DNAase
4) Fragments some organelles such as golgie apparatus by affecting its matrix protiens



Bcl-2 Family they catch the death signal that comes from the extracellular environment or soluble factors that surround the cells

Bcl-2 family share the BH3 domain (more important) and a transmembrane domain(absent in some types)

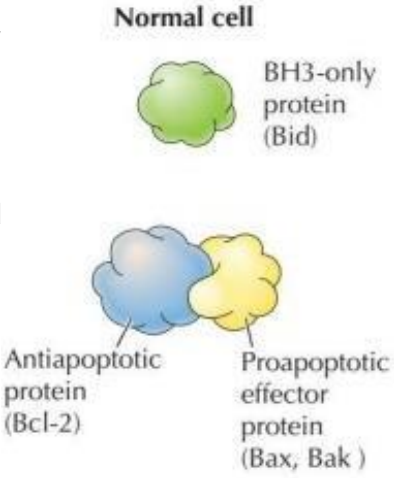


Mechanism of action and classification

- There are three classes of Bcl-2 according to their domains and apoptotic effect:
1. Anti-apoptotic proteins
2. Proapoptotic proteins:
   - *Multi-domain*
   - *BH3-only domain*

The last protein which only contains the TM and BH3 domains called (BH3-Only Protein) and its apoptotic protein it induces apoptosis, in the normal cell its inactive, the other two proteins one of them is proapoptotic and the other is antiapoptotic. The proapoptotic is inactive by the antiapoptotic they are connected to each other and its inactivated this is the normal scenario.

Once a death signal arrives BH3-Only protein is induced only, then it forms a complex with the antiapoptotic and leaves it inactive, and the pro-apoptotic proteins will be free and activate apoptosis.



Internal pathways that induces apoptosis

the presence of permanently damaged DNA stimulates the internal pathway. This leads to apoptosis because keeping damaged DNA can lead to dangerous effects (e.g. disease, cancer).

How is the DNA damages detected?

DNA damage is detected by ATM which activates Chk2. Chk2 then activates p53 by phosphorlyation.
Now, the activated P53 will bind to a certain region on the DNA (puma,noxa,p21,dki,bax) and activates the expression of a group of genes that code for apoptosis

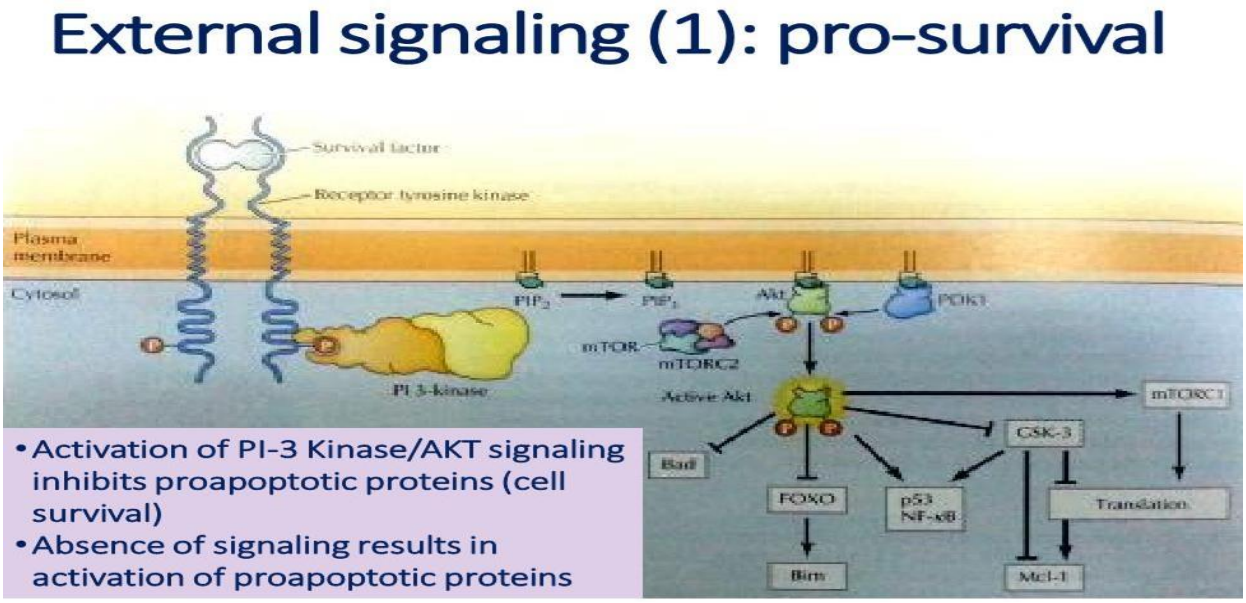
External Pathway

What are the signals that makes the cell to induce apoptosis,and how will it coordinate between signals that induce death and signals that induce survival.

1)Pro-Survival Signals

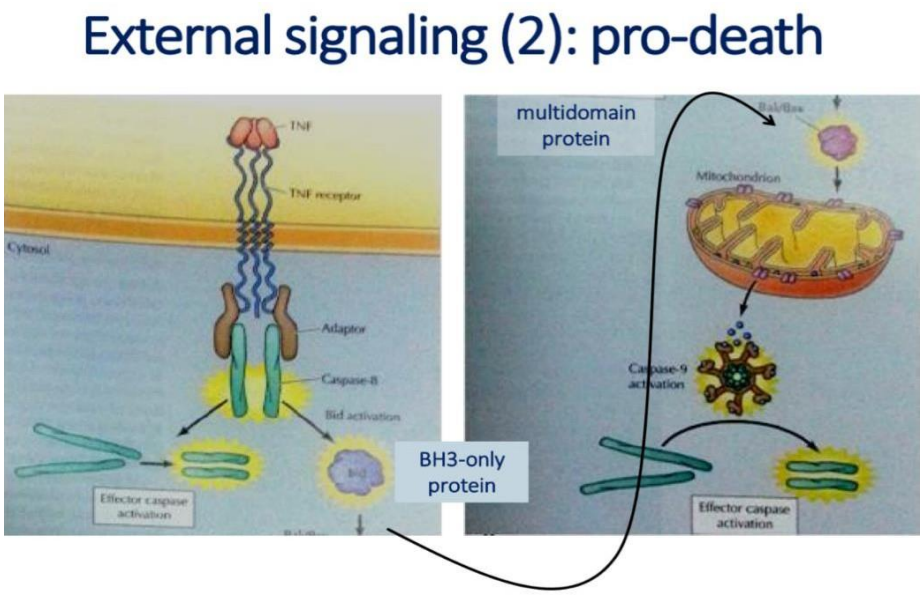
The cell has more than one pathway(BI3-AKT,Ras-Raf/MeK erk ,tumor necrosis factor pathway that eventually causes cell death)

The pathways involved in cell survival will be inhibited,there is no growth factors,no binding with the receptor so the pathway is stopped.



The PIP2-Akt pathway is stopped, no growth factor the PI 3-Kinase wont and Akt wont be activated,no activation of akt no inhibition of bad,the target of Akt inhibit its target so the akt is not active the target proteins will be activated and they're involved in apoptosis.

2)Pro-death Signals



Pathways that induce apoptosis e.g Tumor necrosis pathway

They need to be activated

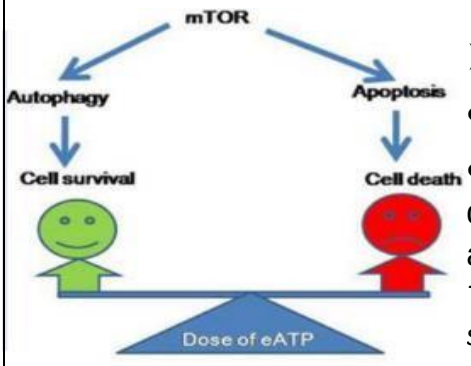
The Tumor Necrosis Factor (TNF) is an external death signal that reaches the cell bind to a TNF receptor which then binds to an adaptor protein that activates caspase-9. The activated caspase then activates BH3-only protein it which then activates the Bax protein to form oligomeric structure which enters the mitochondria releasing the cytochrome c. Then the cytochrome c binds to an adaptor protein which then binds to caspase 9 and it activates caspase 3 resulting in apoptosis.

So to sum it up,we have pro-survival and pro-death signals the pro-survival pathways will be inhbited and pro death signals will be activated that normally get inhibited inducing apoptosis.

Autophagy

Autophagy is not apoptosis

- What is Autophagy?
  - Autophagy is a rescuing process that acts as a final attempt for correction and repair before the cell loses hope and apoptosis takes place.
- What are the advantages of autophagy?
  - When the cell lacks molecular machinery (ex: proteins) of apoptosis; autophagy gives the cell the needed time for preparation of what's required for apoptosis.
  - It gives the cell a chance to repair the damage in it before it undergoes apoptosis



➢ <u>The balance between autophagy and apoptosis:</u>

- Autophagy might take place and then be followed by apoptosis
- Or, autophagy can occur alone and get rid of the damage in the cell without having to be followed by apoptosis

To sum up, different changes occur to maintain the balance in the cell. It may choose cell survival and save itself through autophagy or it may choose to do apoptosis.

This sheet was written in a bad timing, sorry for any mistakes