CYTOLOGY

بسم الله الرحمن الرحيم

MID (last lecture) – Lecture 16

Cell Renewal & Death

﴿ وَإِن تَتَوَلَّوْا يَسْتَبْدِلْ قَوْمًا غَيْرَكُمْ ثُمَّ لَا يَكُونُوا أَمْثَالَكُم ﴾

اللهم استعملنا ولا تستبدلنا

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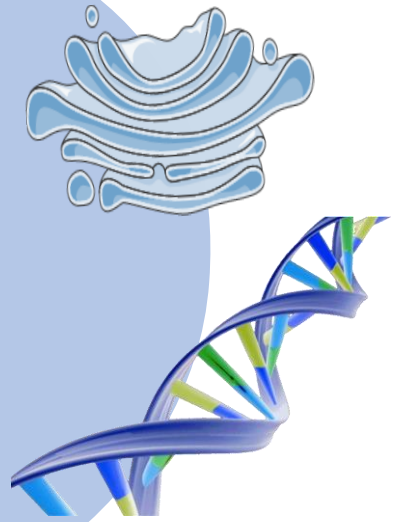

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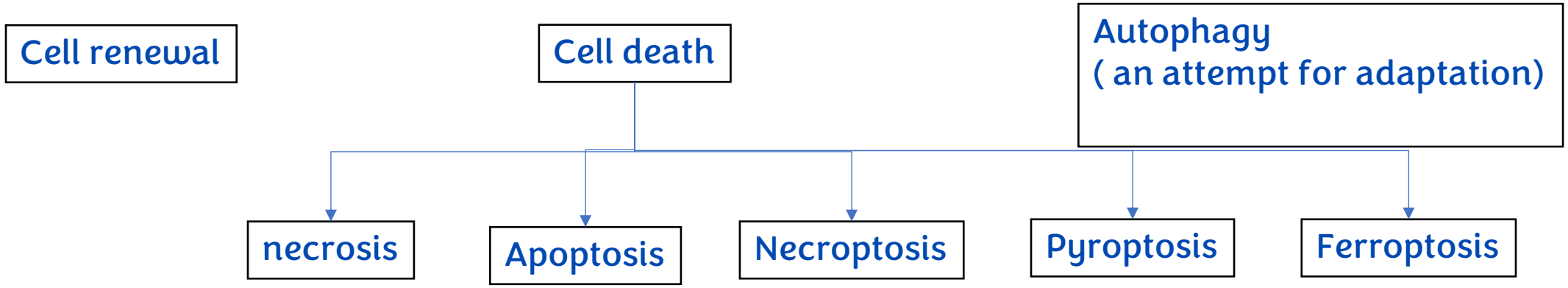

- Regulators
- Pathways:

Extrinsic Apoptosis (Death Receptor Pathway)

Intrinsic Apoptosis (Mitochondrial Pathway)

Lecture 10: Cell renewal and death

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School of Medicine

Second year, Second semester, 2024-2025

Cell renewal

- Most types of differentiated cells are no longer capable of proliferation.
- If these cells are lost, they are replaced by the proliferation of less differentiated cells derived from self-renewing stem cells.
  - Hematopoietic stem cells (>100 x 10^9 blood cells are lost daily)
  - Intestinal epithelial cells

Stem cells that produce blood

- Other types of differentiated cells retain the ability to proliferate as needed to repair damaged tissues.
- These cells enter the G_0 stage of the cell cycle but resume proliferation as needed to replace cells that have been injured or died.
  - Examples: fibroblasts, endothelial cells, Smooth muscle cells, epithelial cells (liver)

They resume proliferation when they get appropriate signals.

In cell renewal old cells are replaced with new cells (that differentiated from stem cells) or by proliferation of progenitor cells, maintaining structure and function.

Renewal is carried out in case of: cells losing their ability to proliferate, or in case the tissue is damaged, or if some certain cells have received specific signals.

Programmed cell death

Programmed cell death is timed, well regulated process.
It is timed occasionally, such as in embryogenesis, where cells appear then cleaved in later stages.
For example: the development of the hand ,in the beginning
hands are formed as a one mass with no fingers then some cells die within this mass and proteolysis
Is carried out resulting in fingers formation.

- Most cell death occurs by a process of programmed cell death either normally or due to cell/DNA damage.
- It plays a key role both in the maintenance of adult tissues and in embryonic development.
  - Renewal of >100 x 10^9 blood cells a day
  - Elimination of nerve cells with a faulty connection
  - Elimination of damaged and potentially dangerous cells
    - Cells with DNA damage
    - Virus-infected cells

Here the cell death occurs due to an induction

- It is a normal physiological form of cell death with a distinct process known as apoptosis ("leaves falling down").

The Greek meaning of Apoptosis

Necrosis

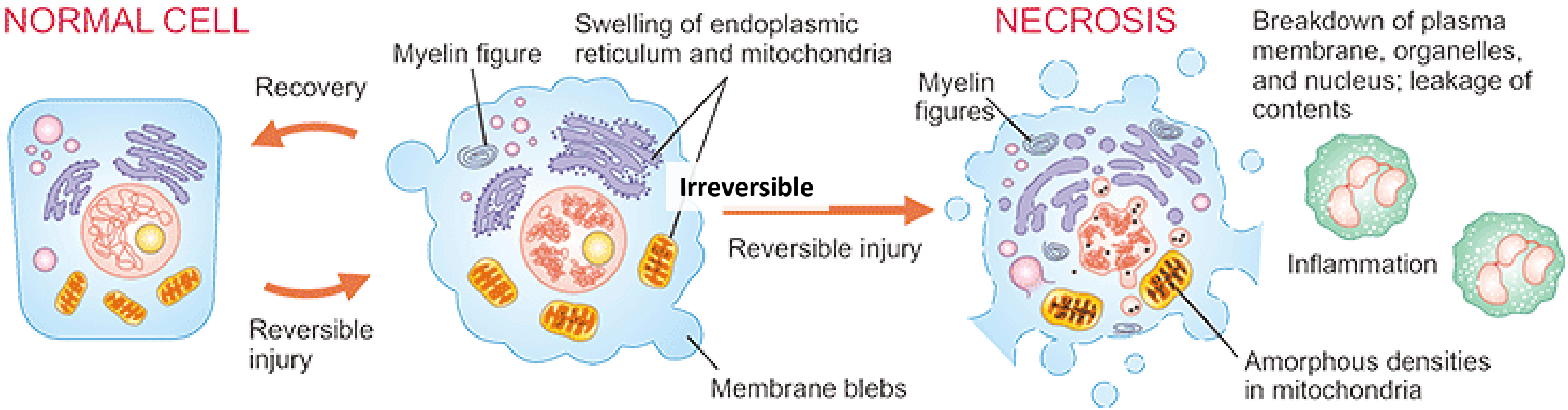
It occur due to something from outside, not within the cell

In necrosis : plasma membrane damage → cell becomes leaky → fluids get in and cellular Contents leak out → resulting in inflammatory response.

For an example: Cell in core of tumor mass experience lack of nutrients, and are damaged , therefore necrosis takes place.

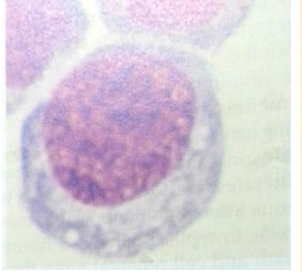
- The accidental death of cells that results from an acute injury.
- Cell necrosis results in membrane damage, enlargement of cells, release of intracellular contents, and inflammation.

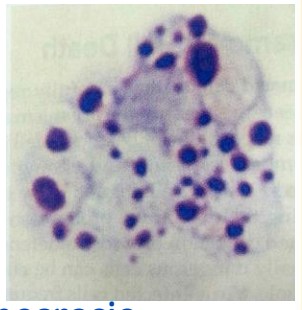
Necrosis will be known by swelling of the cell



Apoptosis

- Fragmentation of chromosomal DNA
- Chromatin condensation
- Nuclear fragmentation
- Cell shrinkage
- Cell fragmentation (apoptotic bodies)
- Phagocytosis by macrophages and neighboring cells (“eat me” signal)



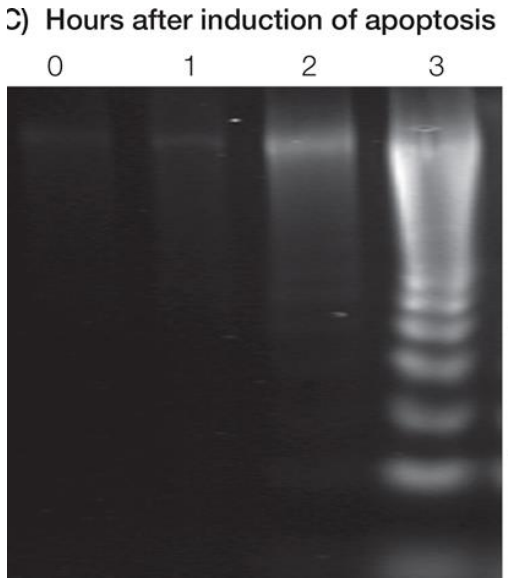


the nuclear skeleton which is composed of Lamin will get fragmentated .

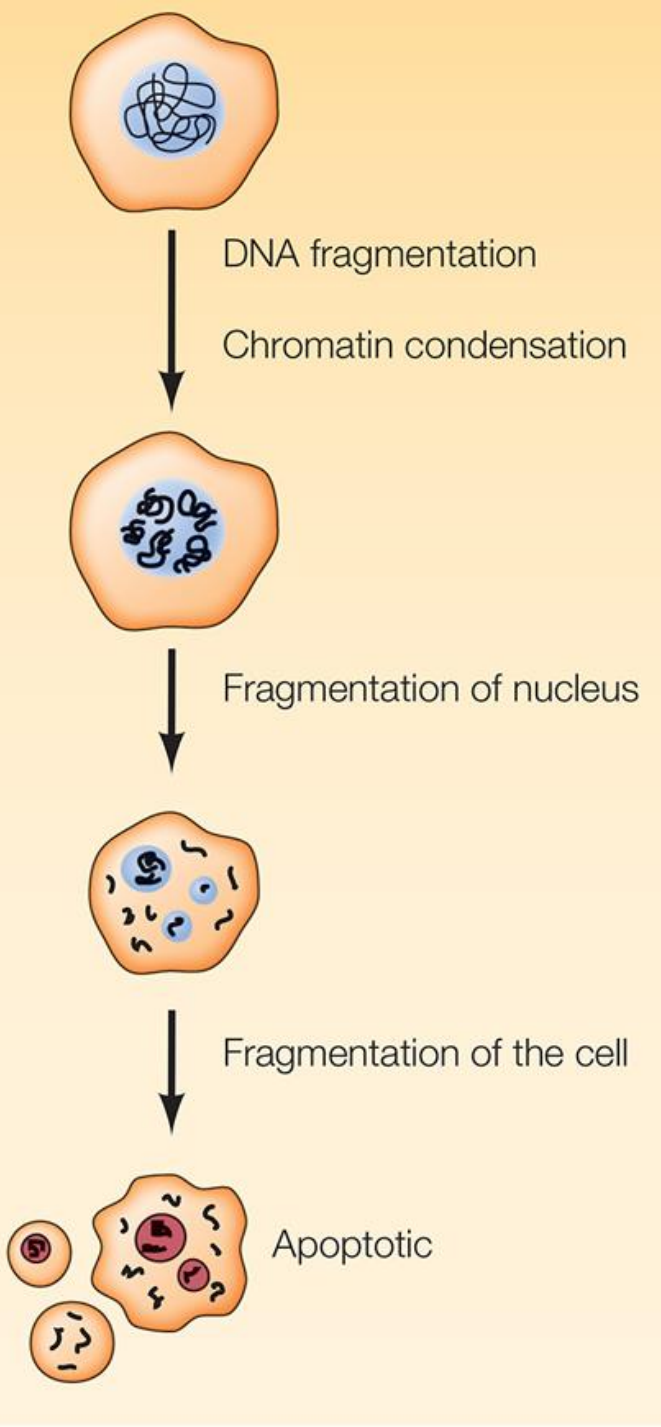
the most important difference between apoptosis and necrosis.

At the end , macrophages will clear up the region where apoptosis occurred by phagocytosis.

We can indicate apoptosis by the chromosomal DNA fragmentation through a technique called gel electrophoresis, which separate the fragments based on its size, each band contain fragments of similar MW.



Courtesy of Ken Adams, Boston Univer



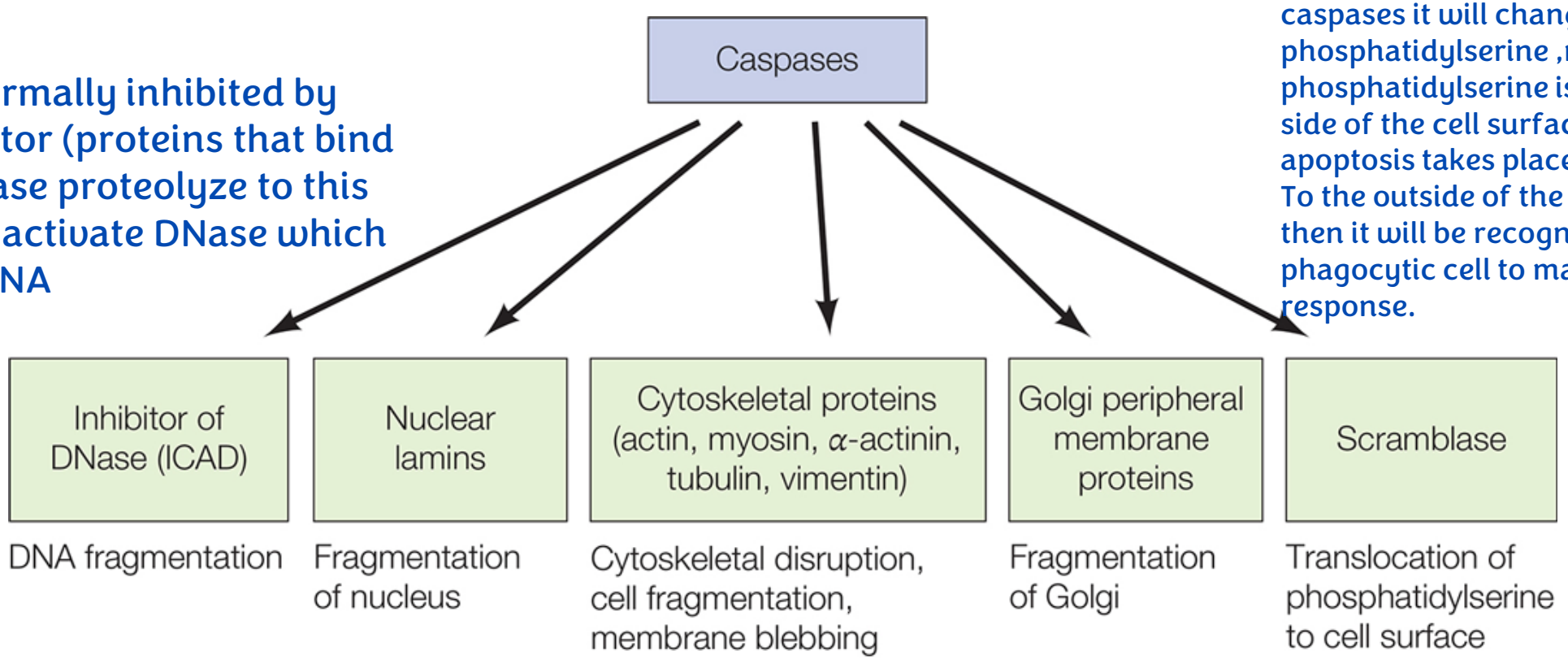
How fragmentation occur?
By enzymes that degrade proteins (caspases) , which have different targets

Caspases: the culprits of apoptotic actions

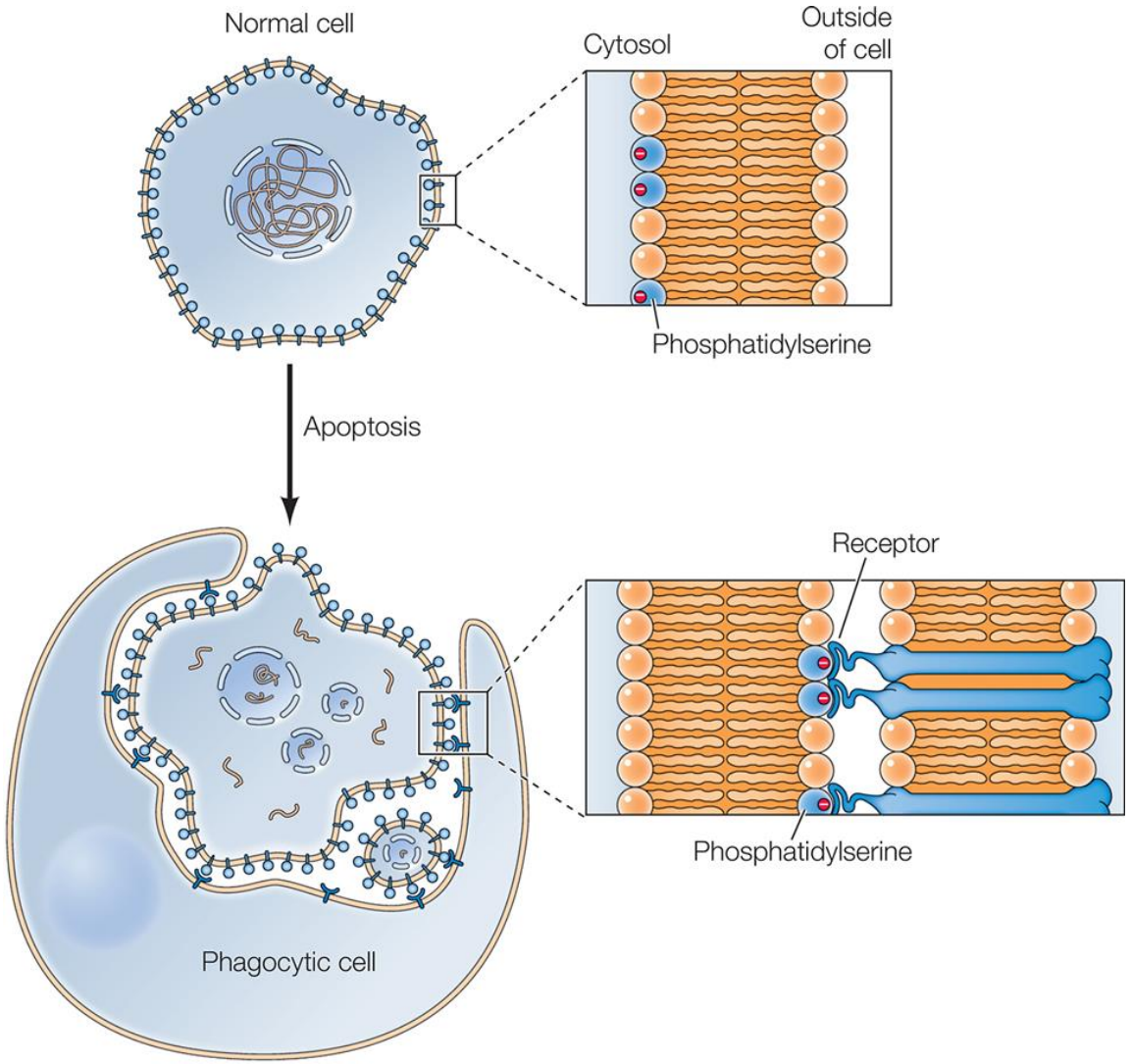
- The caspases are the ultimate effectors or executioners of apoptosis cleaving more than 100 different target proteins (know specific examples).

DNase is normally inhibited by some inhibitor (proteins that bind to it) , caspase proteolyze to this inhibitor to activate DNase which fragment DNA

scramblase, when it gets activated be caspases it will change the position of phosphatidylserine ,normally phosphatidylserine is on the cytosolic side of the cell surface but when apoptosis takes place it will get exposed To the outside of the cell by scramblase then it will be recognized by the phagocytic cell to make inflammatory response.



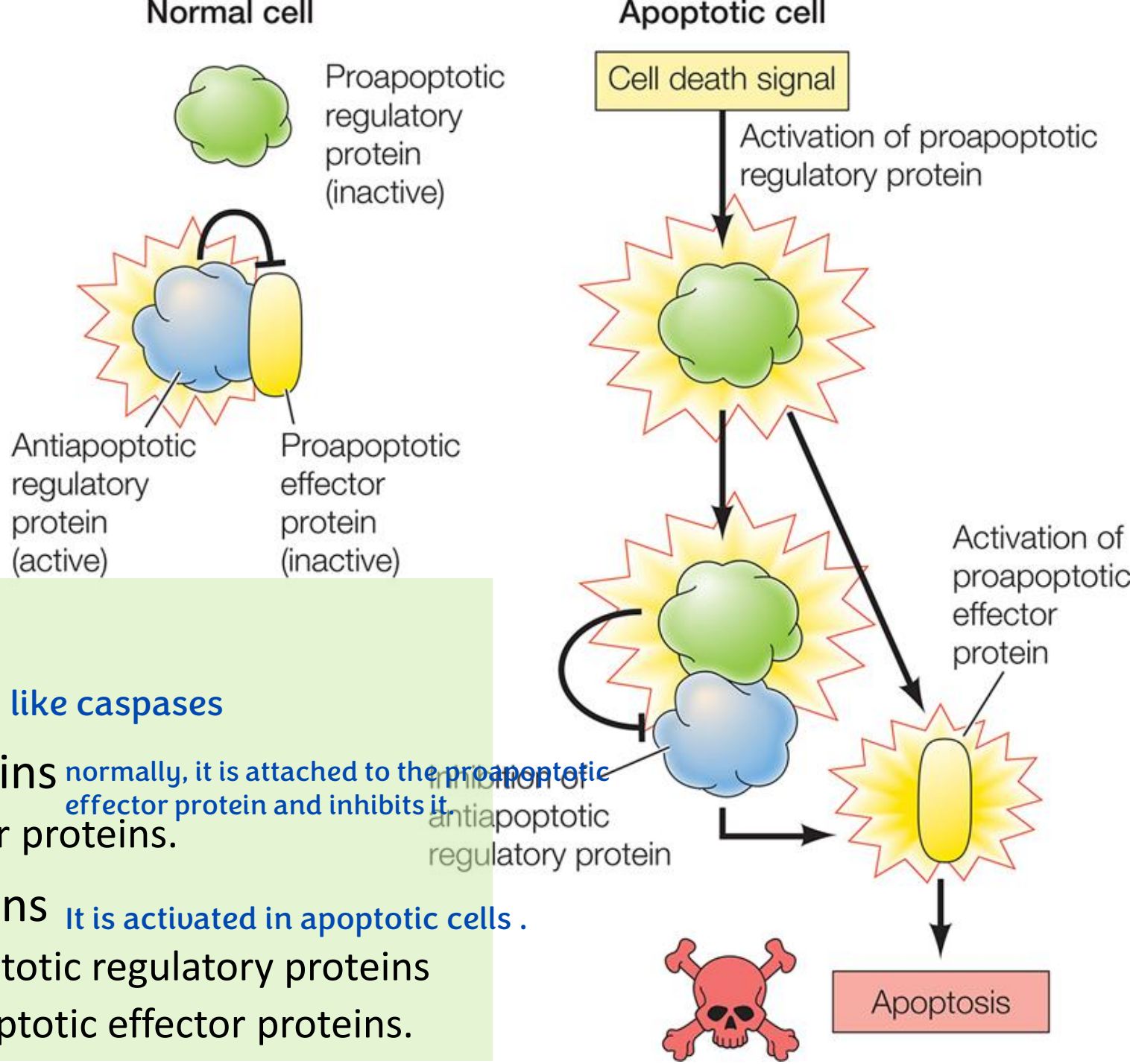
Role of phosphatidylserine



- Normally, PS is expressed on the inner leaflet of cells.
- During the initiation of apoptosis, PS is flipped to the outer leaflet.
- It is then recognized by phagocytic cells.

It is an early marker of apoptosis.

The regulators



The Bcl-2 family

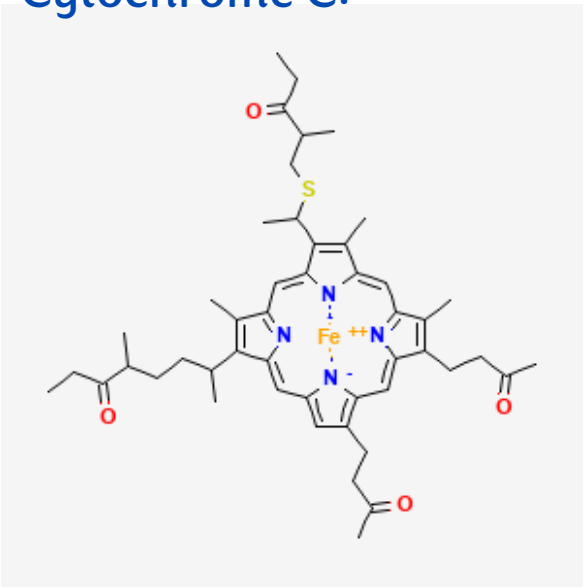
- The proapoptotic effector proteins like caspases
- The antiapoptotic regulatory proteins normally, it is attached to the proapoptotic effector protein and inhibits it.
  - They inhibit the proapoptotic effector proteins.
- The proapoptotic regulatory proteins It is activated in apoptotic cells .
  - They bind to and inhibit the antiapoptotic regulatory proteins
  - They allow the release of the proapoptotic effector proteins.

The molecular activation of apoptosis

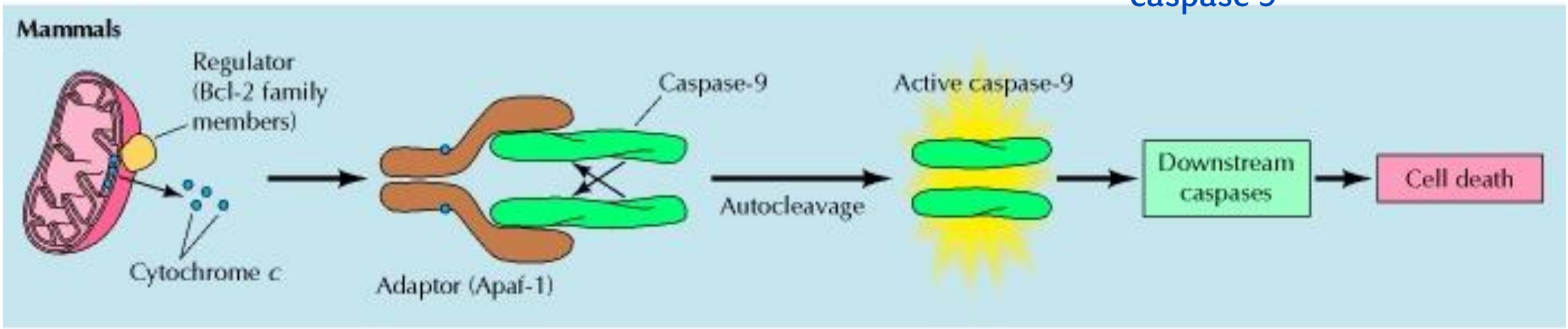
note : the names of proteins are not included unless it is written in slides or the doctor explained it.

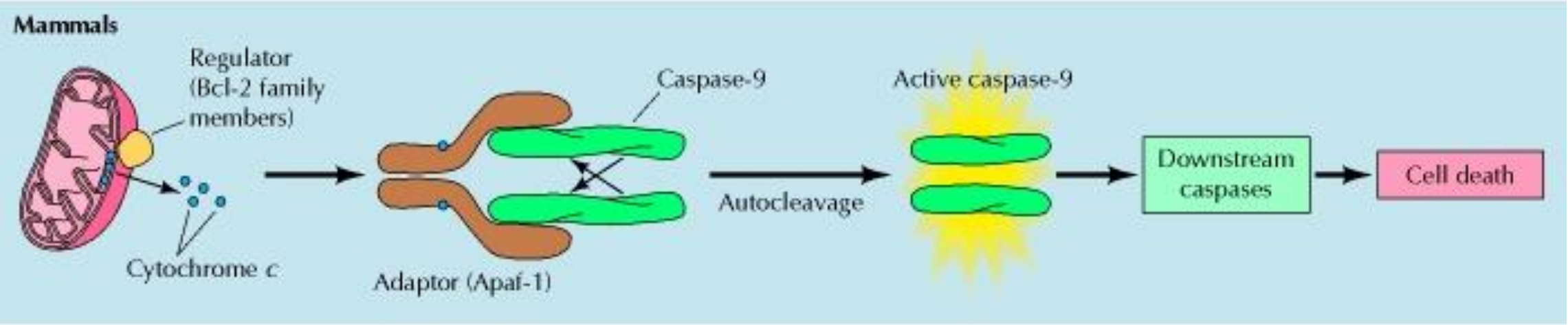
- Activation of the apoptotic pathway involves stimulation of the proapoptotic effector proteins that oligomerize to form pores in the mitochondrial outer membrane.
- Cytochrome c, which exists in the intermembrane space, is released and forms the **apoptosome complex** with caspase 9 and Apaf-1.

Cytochrome C:



Apoptosome complex:
- cytochrome
- Apaf 1
- caspase 9





1. Apoptosome complex form a pore in the outer mitochondrial membrane.
2. This lead to Cytochrome C leakage to cytosol.
3. Cytochrome C binds to Apaf-1 & Caspase-9.
4. Activated Caspase-9 work in different Protein targets.
5. The modification on these proteins facilitate the cell death.

Pathways of apoptosis

Apoptosis is stimulated through two different pathways:

- Intrinsic pathway: simulated by DNA damage, viral infection, and cell stress such as growth factor deprivation

the signal comes from within the cell:
cellular damage, specifically DNA damage

- Extrinsic pathway: stimulated by signals from other cells

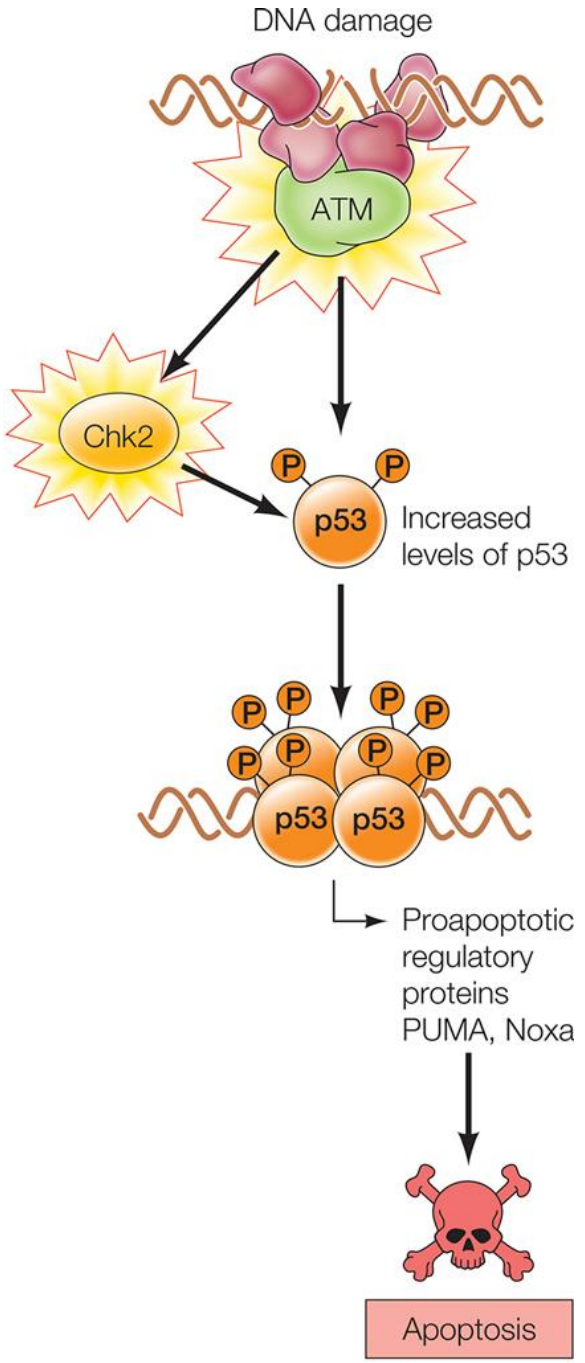
through a variety of mechanism that involve interaction of extrinsic factors with the cell and transduction of the signal to the inside of the cell causing cell death.

They differ in their involvement of Bcl-2 family proteins and in the identity of the caspase that initiates cell death

Intrinsic pathway
DNA damage

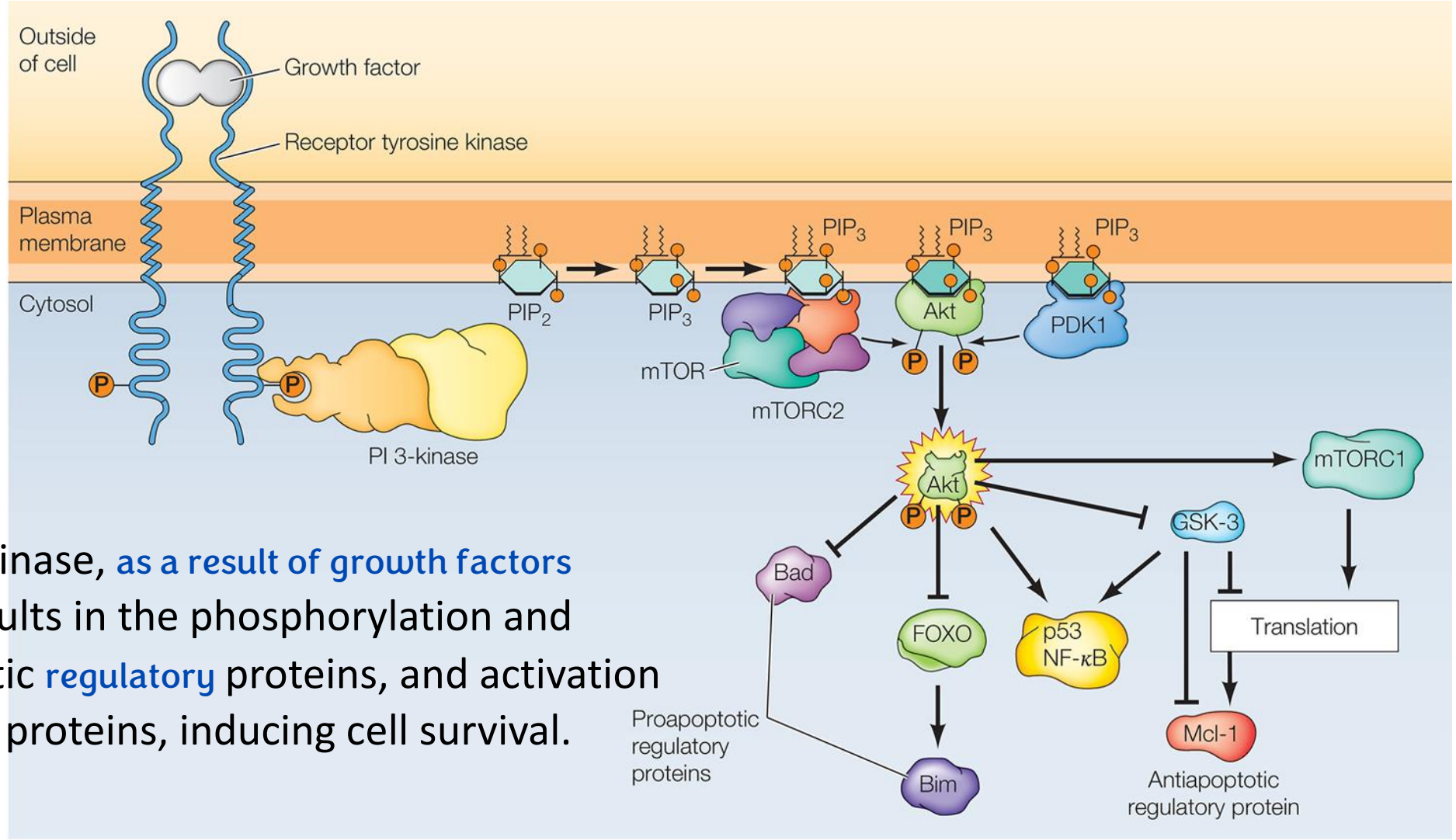
- DNA damage (damage in the phosphodiester bonds between nucleotides causing DNA breakup) leads to activation of the ATM protein kinase, which leads to the phosphorylation, stabilization, and increased levels of p53.
- Afterwards, p53 could either stimulate the repair of the damaged DNA, or–in our case– the transcription factor p53 then activates the transcription of genes encoding proapoptotic regulatory proteins driving cell death when the damage is too severe to be repaired.

  Those transcribed regulatory proteins would proceed to form the spores on the outer mitochondrial membrane leading to the leakage of cytochrome c molecules leading to the formation of the apoptosome complex…



Intrinsic pathway Growth factors

This slide gives an overview on how certain growth factors would inhibit cell death, clearing that absence of these growth factor in some cases would lead to cell death



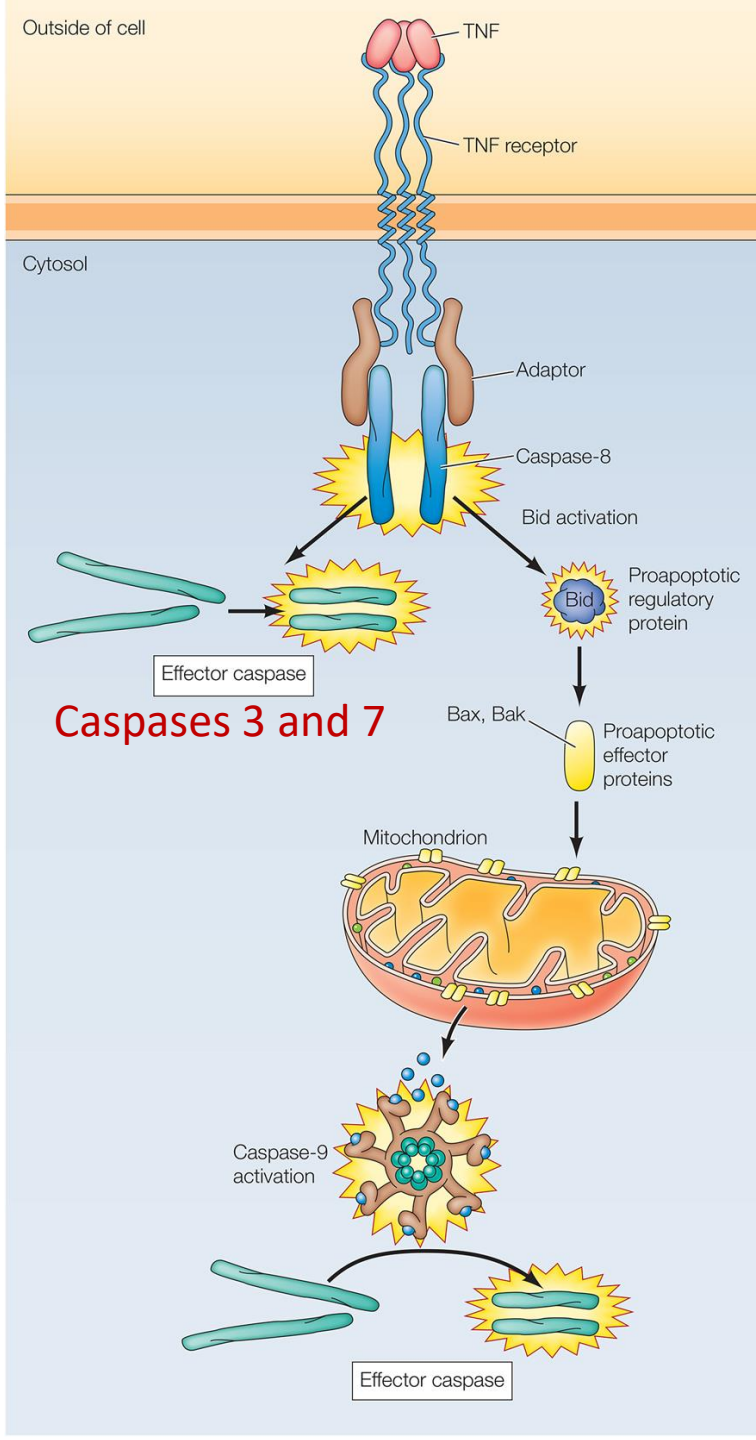
Activation of Akt by PI-3 Kinase, as a result of growth factors binding to their receptors results in the phosphorylation and inactivation of proapoptotic regulatory proteins, and activation of antiapoptotic regulatory proteins, inducing cell survival.

Extrinsic pathway

The tumor necrosis factor (TNF) signaling

- TNF binds to the TNF receptor such as Fas inducing apoptosis in a variety of cell types like:
  - Apoptosis induced by activation of Fas is responsible for killing target cells of the immune system, such as cancer cells or virus-infected cells, as well as for eliminating excess lymphocytes at the end of an immune response.
- Receptor activation leads to the activation of caspase 8, which either activates effector molecules such as (caspases 3 and 7) or proapoptotic regulatory proteins including, ultimately, caspase 9 (for signal amplification purposes).

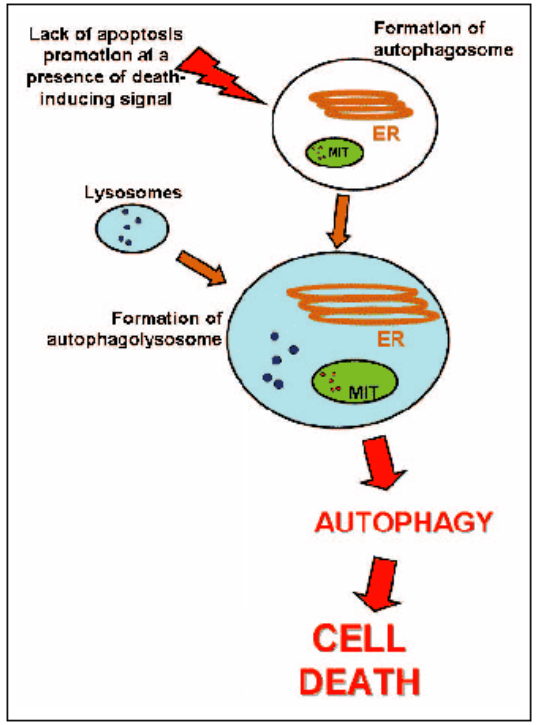
(amplification is needed in case the activation of caspases 3 and 7 alone isn't sufficient for cell apoptosis)

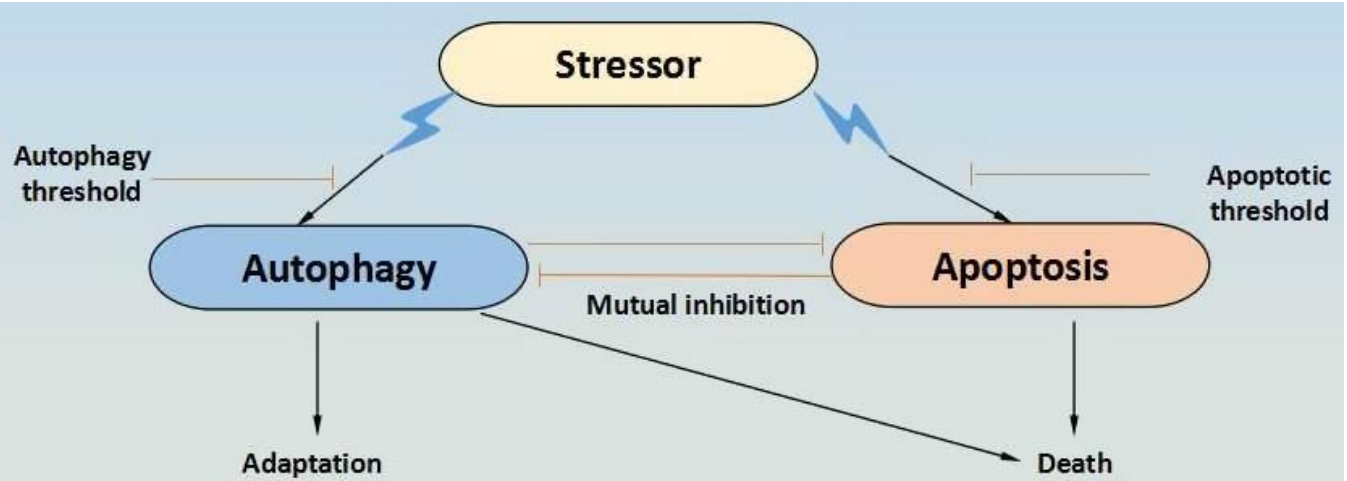


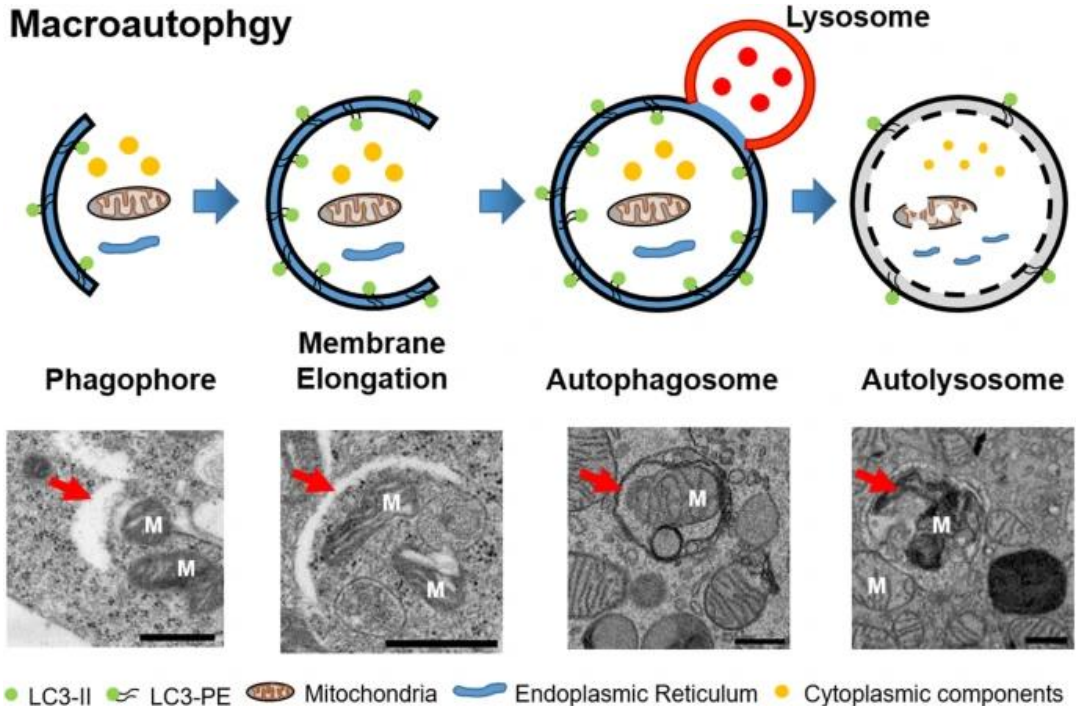
Autophagy (cell self-eating)

An alternative mechanism of apoptosis

- Autophagy is caspase-independent and is mediated by mTOR signaling.
- The dying cell has accumulating lysosomes.
- Autophagy and apoptosis inhibit each other.







Autophagy is a caspase independent alternative pathway that the cell undergoes in cases such as nutrient deprivation when apoptosis could be avoided, because the cell's ultimate goal is survival. During autophagy, the cell's accumulating lysosomes fuse with vesicles that form around organelles intended to be digested for providing energy, forming autophagosomes followed by digestion.

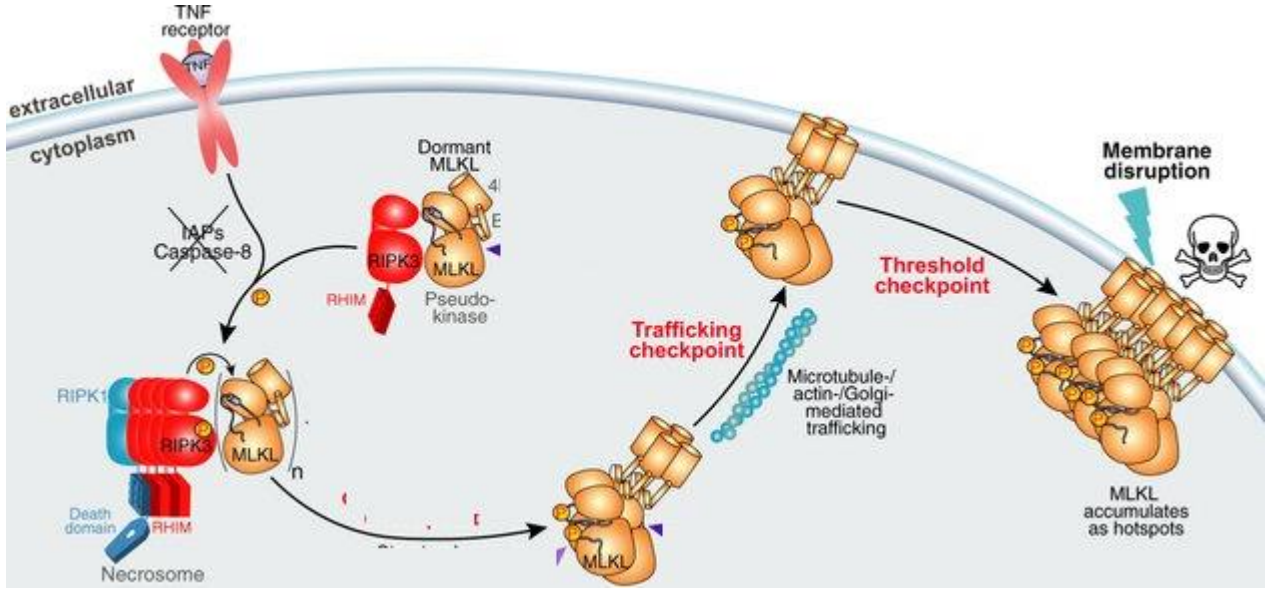
The process is mediated by mTOR signaling as it senses the presence or absence of nutrients. Molecules that induce autophagy inhibit apoptosis, which provides a chance for cell survival.

Necroptosis

It is induced by damage from outside the cell like necrosis, but regulated like apoptosis.

- Necroptosis, like necrosis, results in the extracellular release of intracellular substances triggering an immune response.
- Unlike necrosis, necroptosis:
  - is triggered by specific stimuli such as bacterial infection, DNA damage, or TNF signaling and
  - is executed by a specific molecular mechanism.

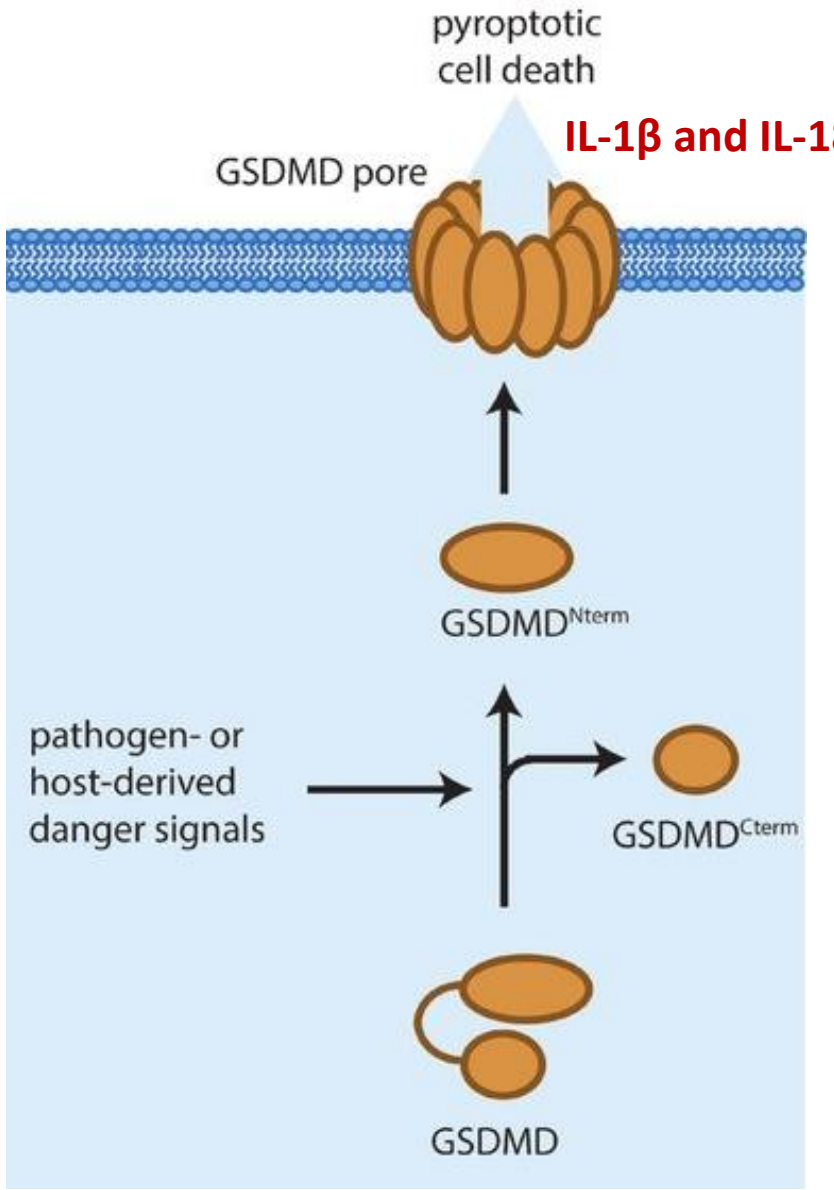
- The protein MLKL assembles into an oligomeric pore in the plasma membrane allowing for a rapid flux of ions into and out of the cell, causing cell swelling and rupture.
- The immune response facilitates the attack.



Pyroptosis

pyro (fire/fever) and ptosis (to-sis, falling)

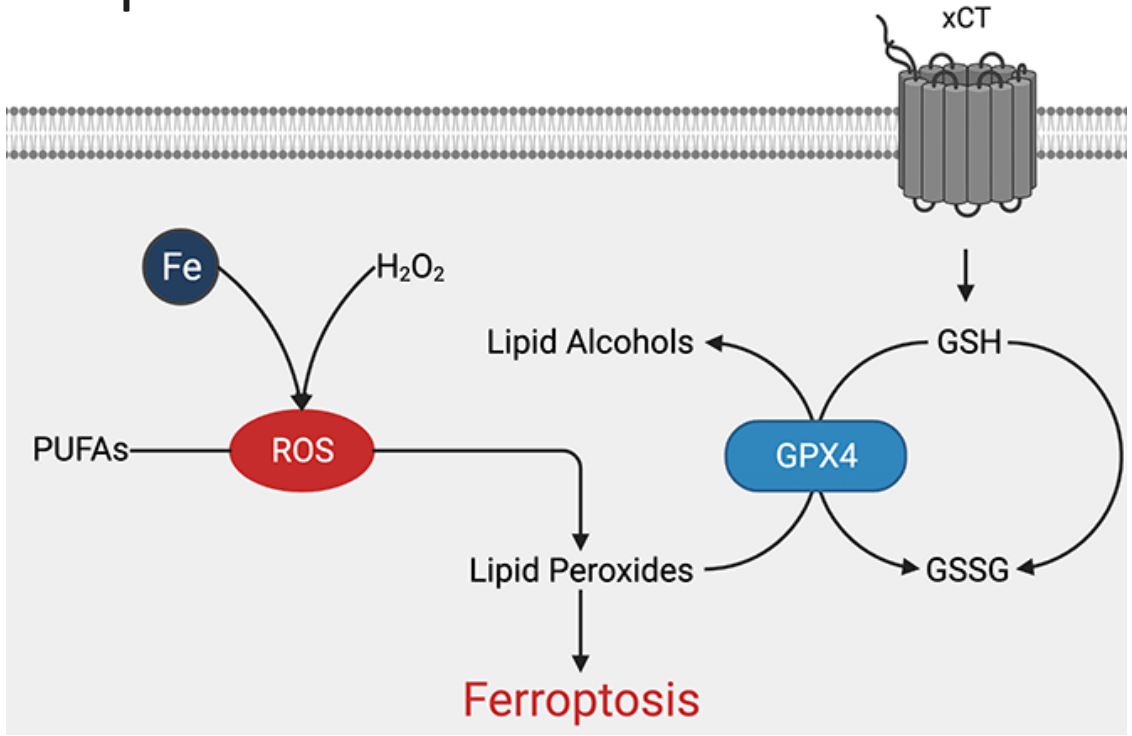
- Pyroptosis is a form of cell death that is triggered by proinflammatory signals and associated with inflammation.
- It is seen primarily in inflammatory cells such as macrophages.
- It is induced by specific stimuli such as microbial infection, executed by specific pyroptotic machinery, and involves activation and oligomerization of a protein—gasdermin—into a pore complex at the plasma membrane.
- A key feature of pyroptosis is the activation and release of pro-inflammatory cytokines IL-1β and IL-18 through the gasdermin pore, which triggers a particularly active immune response.



This mechanism is stimulated by inflammatory signals that are secreted through complexes formed by the oligomerization of gasdermin proteins after induction by certain stimuli such as microbial infections

Ferroptosis

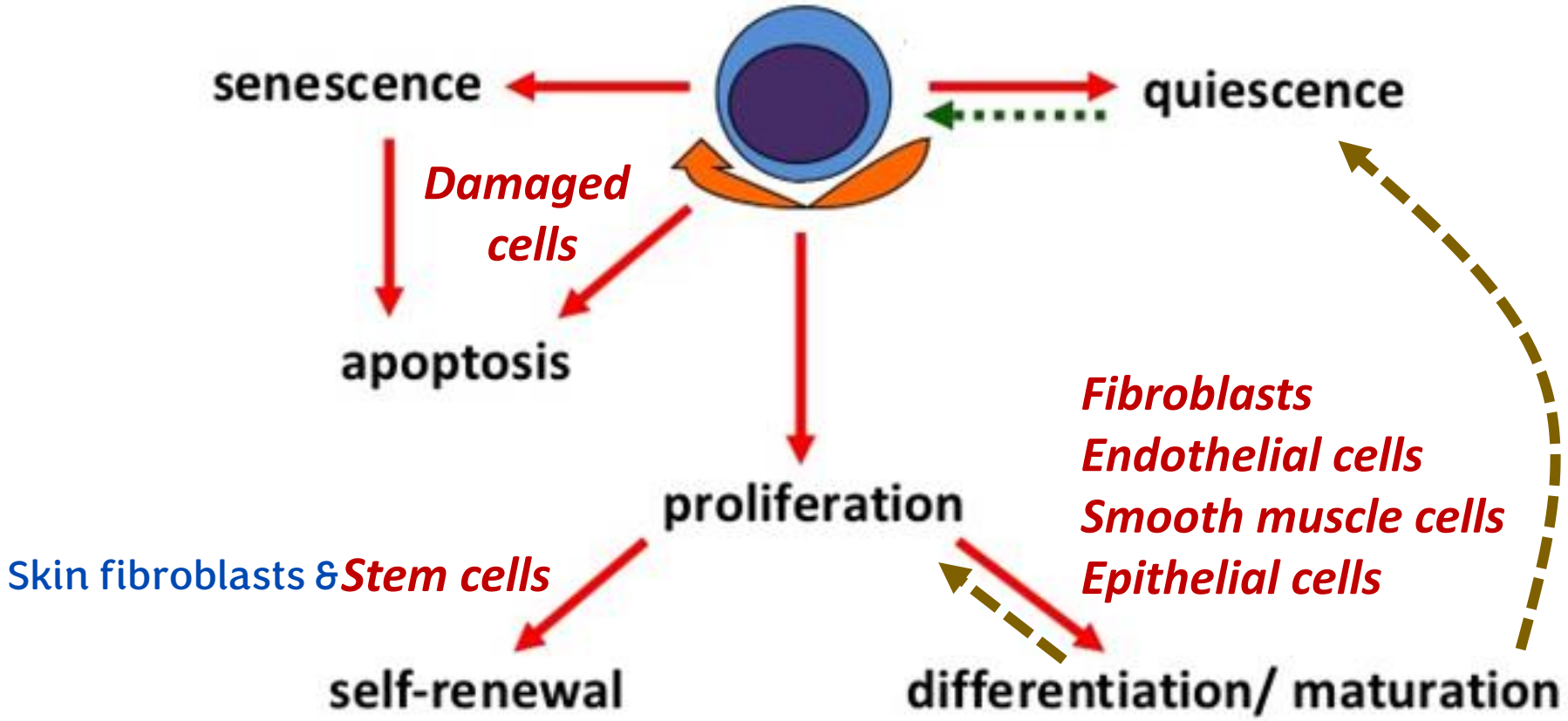
- Ferroptosis is an iron-dependent and oxidative damage-induced cell death that results from iron accumulation and lipid peroxidation, and loss of selective permeability of plasma membrane.
- It involves depletion in the antioxidant enzymes, particularly glutathione peroxidase.



When we have the accumulation of iron inside the cell this increases the oxidizing agents, leading to cell damage, damage to molecules including proteins and plasma membrane and DNA.

Main features of ferroptosis: accumulation of iron and decrease of the antioxidizing agents particularly glutathione peroxidase.

Cell fate


senescence
quiescence
Damaged cells
apoptosis
proliferation
Fibroblasts
Endothelial cells
Smooth muscle cells
Epithelial cells
Skin fibroblasts & Stem cells
self-renewal
differentiation/ maturation

For any feedback, scan the code or click on it.

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1			
V1 → V2			

Additional Resources:

رسالة من الفريق العلمي:

Reference Used:
(numbered in order as cited in the text)

1. The Cell: A Molecular Approach 10th edition

نحيي باسم و محمد[2]
(ربِ اغفر لي ولوالدي وارحمهما كما ربياني صغيراً)

اللهم أحفظ أهلنا في غزة والشام والسودان و في مشارق الأرض ومغاربها, وأعزهم بنصرٍ عاجلٍ من عندك.