4

Pathology

Doctor 2018 | Medicine | JU

◉ Sheet ○ Slides

Done By

Mohammad Abu Zalam

Contributed in the Scientific Correction

Bashar Al Amour

Contributed in the Grammatical Correction

Bashar Al Amour

Doctor

Manar Hajeer

In the last lecture we started discussing the mechanisms of cell injury and now we will continue with that topic.

❖ Endoplasmic Reticulum Stress:

One of the main functions of the ER is protein folding, after the process of translation the protein has to be folded correctly in order to function properly, this is specifically done by proteins in the ER called **chaperons**, they attach to proteins to guide the folding process.

An accumulation of misfolded proteins can happen when the amount of chaperons is very low or non existent, or when the protein concentration is very high to a point where chaperons can't keep up.

Note: misfolded proteins are not functional and are normally ubiquinated (targeted for degradation) and degraded by proteasomes in the cell

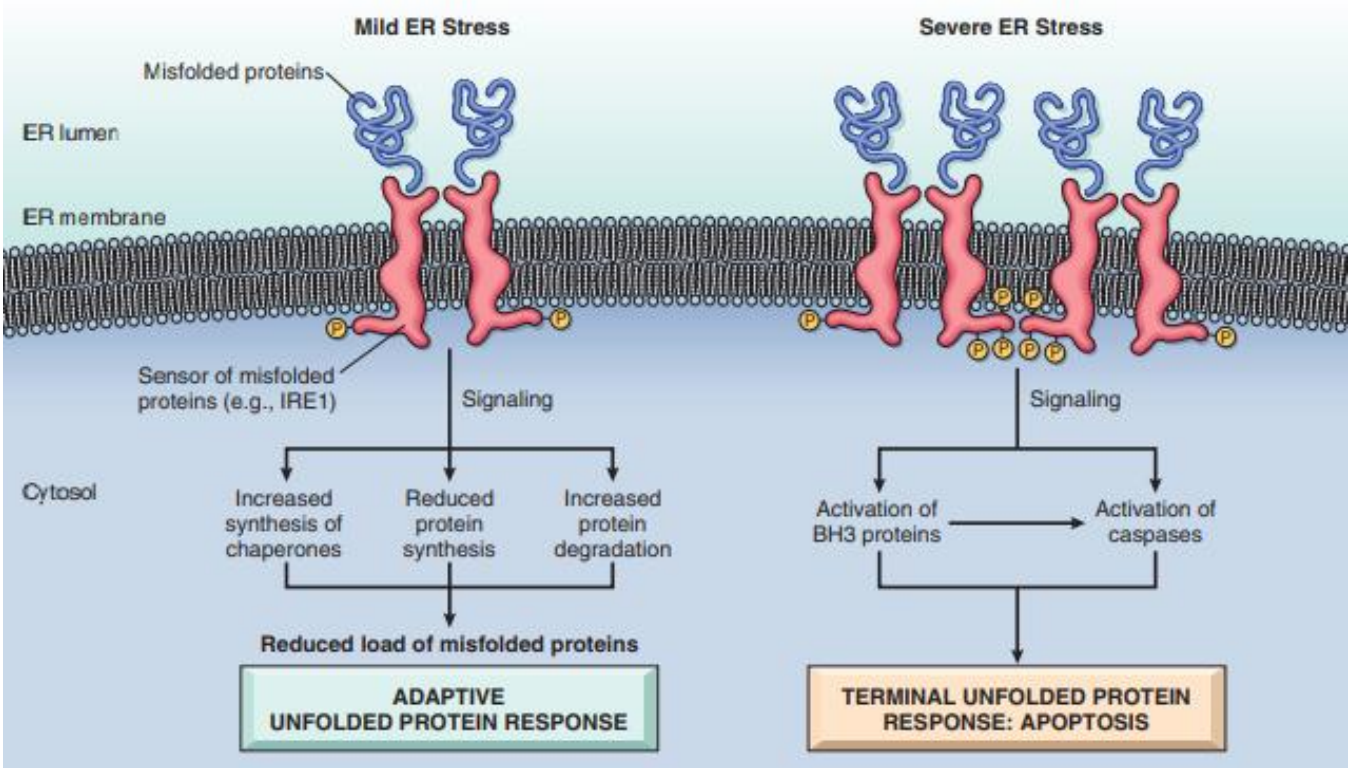
This is where the **Unfolded Protein Response** comes in action, it is an adaptive response where the cell induces the **increase of chaperon production** or the **decrease of protein translation** thus reducing the levels of misfolded proteins in the cell.

When a very large amount of misfolded proteins accumulates and can't be handled by the adaptive response (mechanism failure), the signals that are generated result in the activation of **proapoptotic sensors of the BH3-only family** as well as **direct activation of caspases** (the enzymes responsible for apoptosis), leading to apoptosis by the intrinsic (mitochondrial) pathway.

Note: there are 2 pathways for apoptosis, the intrinsic (mitochondrial) pathway and the extrinsic pathway (death receptor), which will be discussed later

The picture below summarizes what we have discussed

on the Unfolded Protein Response and its failure ☺



❖ Causes of Misfolding:

1. Gene mutations: the protein itself is mutated.

2. Aging: which is associated with a decreased capacity to correct misfolding.

3. Infections: especially viral infections which lead to accumulation of misfolded protein, another example is prion infections such as Mad Cow disease where the mechanism of the disease is misfolded protein accumulation.

4. Increased demand for secretory proteins: a good example on this is Type 2 Diabetes, where there is peripheral resistance for insulin which leads your body to think that there is no insulin thus making the pancreas secrete even more insulin to a point where the chaperons can't even properly fold it leading to an accumulation of unfolded protein which induces cell death and pancreatic damage.

5. Changes in intracellular pH in ischemia and hypoxia: the function of the ER is decreased thus the folding capacity is affected and the accumulation of misfolded proteins is the result.

6. Neurodegenerative diseases: some diseases like Alzheimer's disease, Huntington's disease and Parkinson's disease are caused by accumulation of misfolded proteins in the brain where they induce brain cells apoptosis and degeneration.

Protein misfolding causes diseases by:

- **Deficiency of an essential protein due to degradation:** some essential proteins are degraded or nonfunctional due to misfolding and this can cause diseases by deficiency of this protein, Cystic Fibrosis is an example of this (lack of membrane chloride transporter)
- **Inducing apoptosis of the affected cells:** neurodegenerative disorders [brain cells] (Alzheimer's disease, Huntington's disease and Parkinson's disease) and Type 2 Diabetes [pancreatic cells] are results of this.
- **Improperly folded proteins accumulation in extracellular tissues:** Amyloidosis is an example of this.

Extra info: Amyloidosis is when an abnormal protein called amyloid builds up in your tissues and organs. When it does, it affects their shape and how they work. Amyloidosis is a serious health problem that can lead to life-threatening organ failure

❖ DNA Damage:

DNA damage is another mechanism of cell injury and death where apoptosis could happen if the cell was not able to fix that damage.

What causes DNA damage?

- **Radiation**
- **Chemotherapeutic agents**
- **Intracellular generation of ROS**
- **Mutations**
- **Infections**
- **Extremes of temperature**
- **UV lights**

Normally in the cell, damage to DNA is sensed by intracellular (guard) proteins, which transmit signals that lead to the activation of p53 protein. p53 first arrests the cell cycle (at the G1 phase) to allow the DNA to be repaired before it is replicated (even more accumulation of DNA damage) however, if the damage is too great to be repaired successfully, p53 triggers apoptosis (mainly by stimulating BH3-only sensor proteins).

When p53 is mutated or absent (as it is in certain cancers), cells with damaged DNA survive instead of undergoing apoptosis. In such cells, the DNA damage may result in mutations or DNA rearrangements that lead to neoplastic transformation (conversion of a tissue with a normal growth pattern into a malignant tumor).

❖ Inflammation:

Inflammation is another mechanism of cell injury and death.

What causes inflammation?

- **Pathogens:** such as bacteria, parasites, fungi and viruses
- **Necrotic Cells:** inflammatory cells that clean areas of necrosis cause inflammation
- **Dysregulated immune responses**

Inflammatory cells (neutrophils, macrophages, lymphocytes) use phagocytosis to engulf microbes or dead cell fragments and induce their lysosomal enzymes and reactive oxygen species to kill these microbes and dead cell fragments.

There are 2 common events that happen in cell injury from diverse causes that we mentioned before:

- **Mitochondrial Dysfunction**
- **Defects in Membrane Permeability**

❖ Mitochondrial Dysfunction:

The mitochondria is the energy factory of the cell and any mitochondrial dysfunction that could happen due to hypoxia, toxins or radiation will cause ATP depletion ultimately leading to cell death whether by necrosis or by apoptosis.

What are consequences of mitochondrial damage?

- **Failure of oxidative phosphorylation, ATP depletion**: when ATP is depleted there is no protein synthesis and no sodium-potassium pump function.
- **Abnormal oxidative phosphorylation, formation of ROS:** radicals which will induce cell damage.
- **Mitochondrial permeability transition pores, loss of membrane potential:** the opening of these pores leads to the loss of mitochondrial membrane potential and pH changes, further compromising oxidative phosphorylation
- **Release of cytochrome c leading to apoptosis:** when cytochrome c is released into the cytoplasm it activates caspases leading to apoptosis

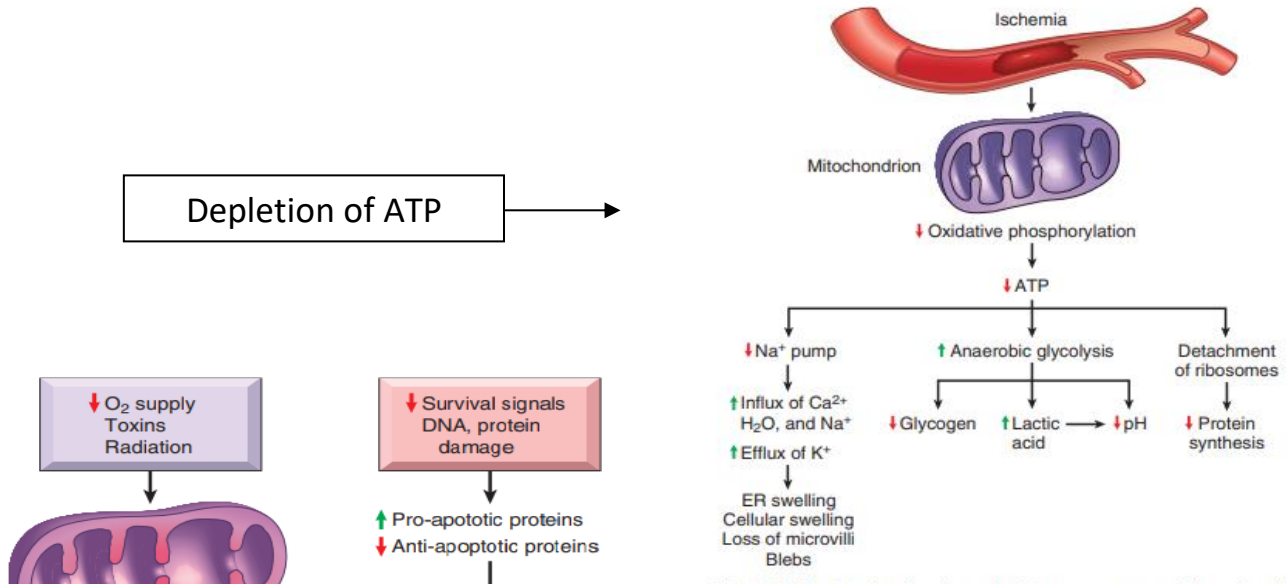


Fig. 2.16 The functional and morphologic consequences of hypoxia and ischemia. *ER*, Endoplasmic reticulum.

Fig. 2.19 Role of mitochondria in cell injury and death. Mitochondria are affected by a variety of injurious stimuli and their abnormalities lead to necrosis or apoptosis. This pathway of apoptosis is described in more detail later. *ATP*, Adenosine triphosphate; *ROS*, reactive oxygen species.

❖ Defects in Membrane Permeability:

When we mention the word membrane we don't just refer to the cellular membrane, we are rather referring to all membranes in the cell such as mitochondrial membrane and lysosomal membranes.

- **Mitochondrial membrane damage leads to decreased ATP.**
- **Plasma membrane damage leads to loss of osmotic balance, influx of fluids and leak of cellular content.**
- **Lysosomal membranes damage leads to leakage of lysosomal enzymes which ultimately leads to cellular damage and digestion.**

Everything we mentioned so far happens in the cell in an interrelated manner at the same time, not sequentially!

The next pages feature a new topic called Apoptosis and Autophagy.

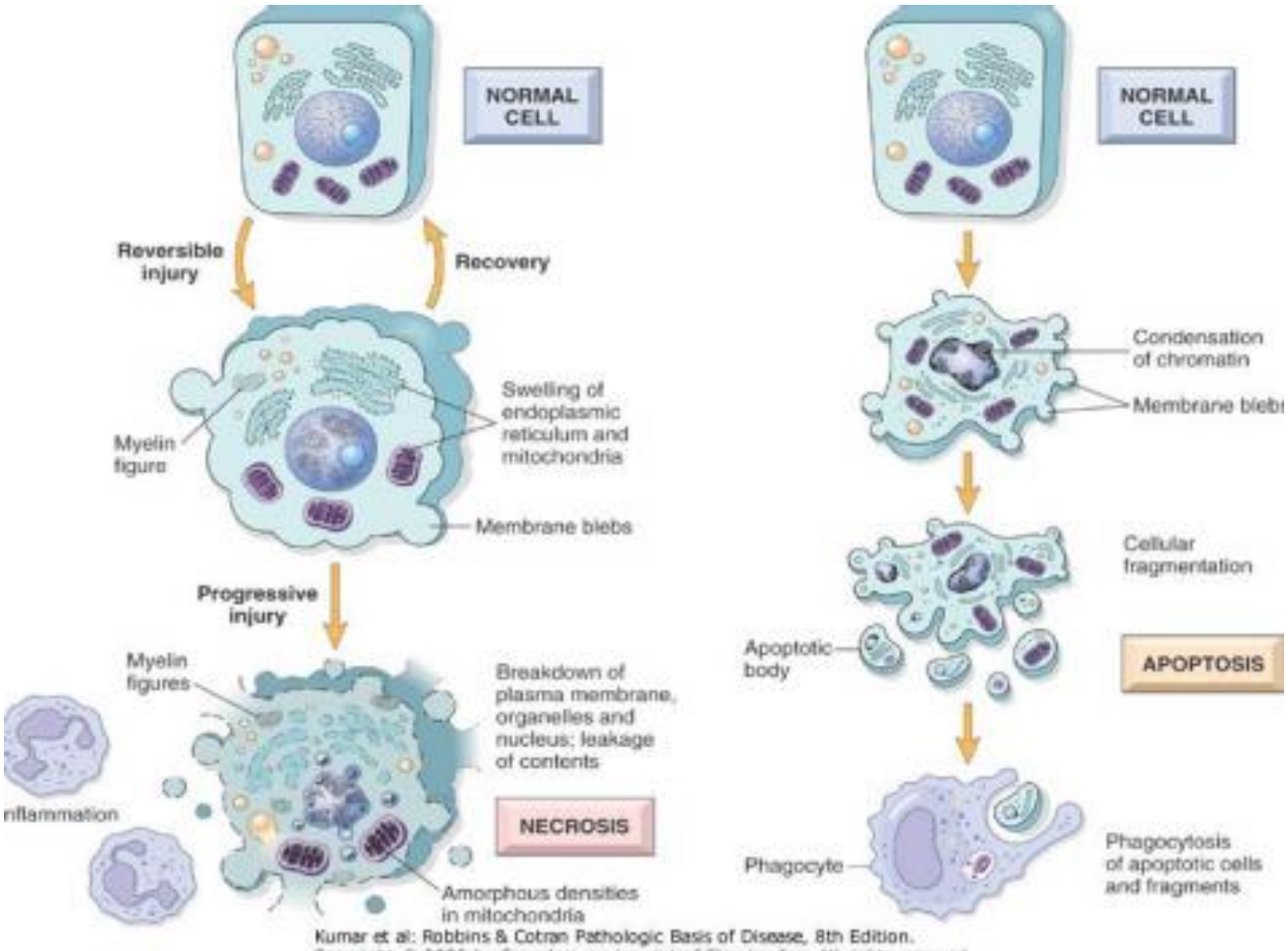
What is apoptosis?

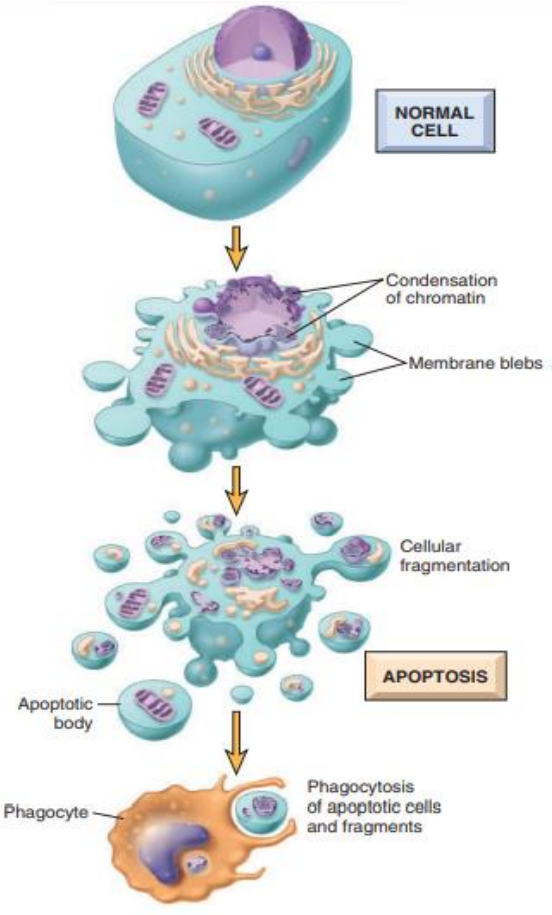
It is a **programmed cell death**, a genetically determined process of cell self-destruction, a pathway of cell death in which cells activate enzymes that degrade the cells' own nuclear DNA and nuclear and cytoplasmic proteins. **It is a highly regulated process at the level of genes and molecules unlike necrosis**.

The dead cell and its fragments are cleared with little leakage of cellular contents, **NO inflammatory reaction** (unlike necrosis)!!

In apoptosis, instead of swelling, shrinking of the cell happens, to a point where it starts falling off and fragmenting, WHILE THE CELL MEMBRANE IS INTACT, which means the cellular content doesn't leak to the outside thus no inflammation happens, but inflammatory cells do still clear the fragments but silently and peacefully.

When insults to the cell are severe and rapid, **necrosis happens**, and when insults to cell are less severe such as DNA damage or misfolded protein accumulation, **apoptosis happens**.



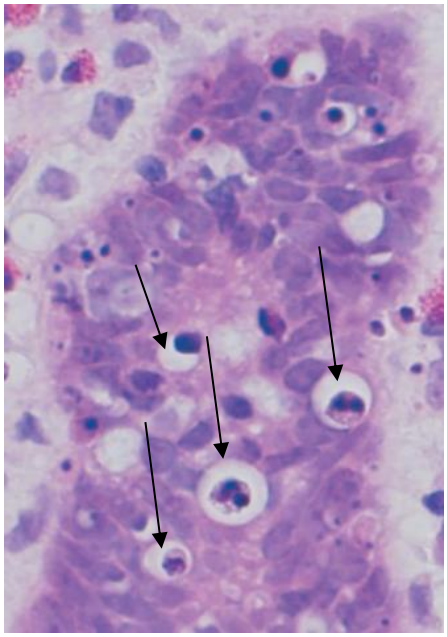


Feature	necrosis	Apoptosis
Cell size	Enlarged(swelling)	Reduced(shrinkage)
Nucleus	Pyknosis, Karyorrhexis, karyolysis	Fragmentation into nucleosome- size fragments
Plasma membrane	**Disrupted**	**Intact**, altered structure, especially orientation of lipids
Cellular content	Enzymatic digestion, may leak out of cell	Intact, may be released in apoptotic bodies.
Adjacent inflammation	**Frequent**	**No**
Physiologic or pathologic role	Invariably pathologic	Often physiologic

This table is very important, be sure to understand the differences between necrosis and apoptosis.

Note: Karyorrhexis is practically the same as fragmentation, but it is reserved for referring to necrosis, it is just a nomenclature difference

apoptosis and necrosis sometimes coexist, and apoptosis induced by some pathologic stimuli may progress to necrosis, like in ischemia.



Notice how the cells shrink in apoptosis leaving behind space (remember lacunae in histology?), the nuclei get fragmented and microphages engulf them soon after.

Causes of apoptosis can be physiologic and pathologic.

Physiologic Apoptosis:

- **During embryogenesis:** apoptosis of cells that get replaced by new organs or tissues in a developing embryo.
- **Involution of tissues upon hormone deprivation:** during lactation and pregnancy, the breasts undergo hyperplasia which is reversible, when lactation stops, the new cell that appeared during hyperplasia disappear by apoptosis. The smooth muscles in the uterus undergo a similar process when they are deprived of estrogen after pregnancy.
- **Steady state population**: as in the apoptosis of aged keratinocytes that are on the surface of the skin for when they get replaced by new keratinocytes for example (If this doesn't happen, an accumulation of keratinocytes will lead to a thick epidermis) the same thing happens to the lining of the colon and intestines.
- **End of function/life**: neutrophils after they do their function at the end of inflammation, they clean the area and get rid of dead cells and microbes and they go through apoptosis
- **Self reacting lymphocytes**: lymphocytes that react to self-antigens causing autoimmune diseases, normally these lymphocytes are gotten rid of through regular apoptosis and shouldn't stay in the body.

Pathologic Apoptosis:

- **DNA damage**: Radiation, Chemotherapeutic agents, UV lights, Hypoxia, Temperature.
- **Accumulation of misfolded proteins**
- **Some infections**: such as Adenovirus, HIV and Hepatitis viruses (they can also induce necrosis at the same time)

Condition	Mechanism of Apoptosis
Physiologic	
During embryogenesis	Loss of growth factor signaling (presumed mechanism)
Turnover of proliferative tissues (e.g., intestinal epithelium, lymphocytes in bone marrow, and thymus)	Loss of growth factor signaling (presumed mechanism)
Involution of hormone-dependent tissues (e.g., endometrium)	Decreased hormone levels lead to reduced survival signals
Decline of leukocyte numbers at the end of immune and inflammatory responses	Loss of survival signals as stimulus for leukocyte activation is eliminated
Elimination of potentially harmful self-reactive lymphocytes	Strong recognition of self antigens induces apoptosis by both the mitochondrial and death receptor pathways
Pathologic	
DNA damage	Activation of proapoptotic proteins by BH3-only sensors
Accumulation of misfolded proteins	Activation of proapoptotic proteins by BH3-only sensors, possibly direct activation of caspases
Infections, especially certain viral infections	Activation of the mitochondrial pathway by viral proteins Killing of infected cells by cytotoxic T lymphocytes, which activate caspases

This table summarizes what we discussed above and it is really important to understand

Good Luck