Chapter 3
IREVERSIBLE CELL INJURY.
CELL DEATH:
NECROSIS AND APOPTOSIS

NECROSIS

Necrosis is a localized death of the cells, tissue or an organ in a living organism. It can be determined by various causal agents: traumatic (the physic or chemic factors), toxic (the bacterial toxins, chemical toxic substances, drugs), neurotrophic (disorders of the trophic function of the central and/or of the peripheral nervous system), vascular (the decreased arterial blood flow in an organ or tissue) and allergic (the lytic action of the immune complexes, antibodies in a sensitized organism).

Microscopic characteristic of the necrosis

a. Nuclear changes:
   - Karyopyknosis - the condensation of the chromatin and the shrinkage of the nucleus
   - Karyorrhexis - fragmentation of nucleus (chromatin).
   - Karyolysis (syn – chromatolysis) - dissolution of chromatin.

b. Modifications of the cytoplasm:
   - Denaturation and coagulation of the cytoplasmatic proteins.
   - Cytorrhexis (plasmorrhexis) – fragmentation of the cytoplasm.
   - Cytolysis (plasmodysis) - liquefaction and hydrolysis of the cytoplasm.

The earlier structural modifications can be easily revealed using electronic microscopy. The precise moment of the cellular death can not be exactly established.

Electronmicrography “Necrosis of the cell, karyopyknosis” (fig. 47).
The nucleus is smaller in volume, its membrane is wrinkled and shriveled. The karyoplasm has an increased electronic density because of the chromatin condensation; the nucleolus is not differentiated. The cytoplasm contains many vacuoles, the mitochondria are tunefacted and homogeneous and the Golgy apparatus is smaller in dimensions.
Karyopyknosis can persist for a long time, after that the nucleus is fragmented into small granules (karyorrhexis) which are scattered into the cytoplasm. The desintegration and dissolution of the nucleus (karyolysis) follows, leaving only a small nuclear shadow. In a day or two the nucleus in the necrotic cell totally disappears.

The intracellular organelles are gradually destroyed. In later phases the cytoplasm is also fragmented and dissolved, forming a homogeneous mass of small granules of amorphous substance called cellular detritus.

Electronmicrography “Focal (partial) necrosis of the cardiomyocyte” (fig. 48).

The sarcoplasm of the cardiomyocyte contains a focus of intracellular organelle destruction and lysis leading to their disappearance and the formation of vacuoles of a low electron density.

Karyopyknosis, karyorrhexis, the destruction of mitochondria, of myofibrils, of the sarcolemma, etc., are all irreversible lesions. According to experimental data, the irreversible modifications in the cardiomyocytes begin after 20 min from the moment of alteration, while the cells of the liver and kidneys are affected after 25-40 min.

**Morphologic patterns of necrosis**

Several patterns of tissue necrosis may be distinguished, reflecting the peculiarities of the macroscopic appearance of the necrotic tissue.

1. Coagulative (or coagulation) necrosis

   It is characterized by the predominance of the densification, denaturation and dehydration (drying) processes of the tissues. The necrotic masses are dry, dense, of a yellowish-white color and may be sharply demarcated from surrounding viable tissue; they do not undergo for an extended time hydrolytic decomposition. This type of necrosis takes place in tissues with relatively few lysosomes to bring about complete breakdown of cellular proteins. The myocardial infarction is prime example.

   Microscopically much of the cellular outlines and tissue architecture can be discerned, cells have a homogenously, compact eosinophilic cytoplasm with loss of cells detail and nuclear changes.

Microspecimen “Necrosis of the contort renal tubular epithelium” (fig. 49).

The epithelial cells of the proximal and distal contort tubules are tumefacted and do not contain nuclei (karyolysis). The cytoplasm is homogeneous, of a pink (eosinophilic) color; the lumen of the tubes is smaller, and even absent in some of the tubes because of the obstructing masses of cellular detritus (plasmorrhexis and plasmolysis). The blood vessels are dilated and hyperemic. The cellular structure of the glomerulus, Henle’s loop and of the collector tubules is unchanged.

Necrotic nephrosis appears as a result of hemodynamic disorders (cortical ischemia of the kidneys) and of the toxic action upon the nephrocytes of chemical substances such as mercury bichloride, ethyleneglycol, etc. Clinically it is manifest by acute renal insufficiency (oliguria or anuria). It is also seen in shock conditions (cardiogenic, traumatic, toxic, bacterial, hemorrhagic, posttransfusional, etc.).

The eventual consequences of necrotic nephrosis are convalescence (the regeneration of the renal tubules and the reestablishment of diuresis) or death as a result of uremia.
Irreversible cell injury

**Microspecimen “Necrosis of the striated muscles (zereus or Zencker’s necrosis)” (fig. 50).**

Among the normal muscular fibers there can be observed necrotic fibers with a tumeffacted cytoplasm, and without nuclei and transversal striation. Some of them are fragmented into homogeneous proteinc blocks of irregular form and various dimensions (plasmorrhesis). Macroscopically necrotic areas have a waxy appearance. It is more frequently seen in skeletal muscles in typhoid fever, exanthematous typhus, especially in the rectus abdominis muscles.

2. Liquefactive (colliquative) necrosis

The prevalence of softening, liquefaction and the autolysis of the dead tissues is observed; the necrotic masses appear semi-liquid, have a soft, flaccid consistency as a result of dissolution of tissue by the action of hydrolytic enzymes. Occurs in tissues that is rich in water and have high lysosomal and lipid content and the processes of hydrolysis are intense. More frequently it is met in the infarcts of the brain and spinal cord (white or gray cerebral softening).

In the nervous tissue the huge lysosomal content in neurons and relative lack of extracellular structural proteins (collagen), leads to rapid liquefaction under action of the lysosomal enzymes.

Macroscopically the cells loose their membrane envelope and become granular eosinophilic and basophilic debris. Tissue architecture is obliterated.

**Macrospecimen “White cerebral softening” (fig. 51).**

The left hemisphere of the brain is deformed. In the subcortical nuclei area and in the occipital region there can be observed necrotic foci of irregular form, soft consistency, composed of a yellowish-white gelatinous mass.

It is seen usually in atherosclerosis and in the hypertensive disease (cerebral ischemic infarct). Clinically, it is manifest by paralysis (monoplegia, hemiplegia, aphasia, etc.). The most frequent consequence of cerebral softening is the formation of a cystic cavity (cystic transformation). If the necrotic site has relatively small dimensions, organizing processes begin, leading to the formation of a glial-connective scar.

**Macrospecimen “Cerebral cyst” (fig. 52).**

On transverse section the brain contains a well delimited cystic cavity, filled with a gelatinous mass. It is formed after the softening and reabsorption of the liquefied necrotic masses in the central portion of the cerebral ischemic infarct site.

3. Caseous necrosis

Is a distinctive form of coagulative necrosis. The dead tissues is soft, has a white-yellow color and a “cheese-like” appearance on gross examination.

Occurs commonly in tuberculosis and some chronic diseases (syphilis, lymphogranulomatosis, leprosy).

Macroscopically the necrotic area appears as amorphous homogenously pink granular debris with loss of cellular integrity. Tissue architecture is obliterated.
Macroscopic “Caseous necrosis of the mesenteric lymph nodes” (fig. 53).
The mesenteric lymph nodes are larger, have a tight mutual adhesion forming packages and conglomerations of dense consistency. On section the lymphoid tissue has a whitish-gray color, resembling dry ewe’s cheese.

Microscopic “Caseous necrosis of the lymph node in tuberculosis” (fig. 54).
Necrotic foci can be observed in the lymph node, which show an amorphous, granular, intensively eosinophilic mass. At the periphery there can be observed nuclear fragments (karyorrhexis); the surrounding tissue contains tuberculous granulomas with Langhans polynucleated giant cells.
The most frequent consequences of caseous necrosis are calcification (petrifying) and encapsulation of the necrotic focus.

4. Gangrenous necrosis:
It represents the necrosis of those tissues that have contact with the outer environment (air, bacteria). It is characterized by the brown-gray or black color of the mortified tissues; the most frequent localization is: extremities, superficial soft tissues, digestive tract, lungs, uterus, urogenital tracts. There can be distinguished dry, liquefactive and gas (anaerobe) gangrene.
The prevalent processes of dry gangrene are drying, densification and shriveling of the mortified tissues.

Macroscopic “Dry gangrene of the foot” (fig. 55).
The tissues affected by gangrene are dry, wrinkled, and mummified as a result of the evaporation or absorption of the water by the normal neighboring tissues. They have a black color and a dense consistency. A well defined delimitation line can be observed between the normal and the affected tissues (demarcating inflammation).
The most frequent causes of the extremity gangrene are: thrombosis or thromboembolism of arteries in atherosclerosis, diabetes mellitus, obliterating endarteritis, burns, freezing, vibration disease, etc. The black color is due to the iron sulphite, which is formed as a result of the interaction of the hemoglobinogenic pigments with the atmospheric air and the hydrogen sulfate produced by the bacteria found in the mortified tissues. During demarcation inflammation, a progressive erosion of the necrotic tissue can be observed which leads to its complete detachment or autoamputation.

Macroscopic “Liquefactive gangrene of the foot” (fig. 56).
The affected tissues are tumefaced, imbued with liquid, of a soft consistency and a bluish-gray or blackish color. There is a smell of putrefaction, and there is no demarcation line.
It is mostly seen in the lower extremities (in diabetes mellitus), in the lungs (as a complication of pneumonia, pulmonary abscesses and infarcts), and in the intestines (in atherosclerosis). It evolves because of the action of putrefaction saprophyte bacteria (Bac. fusiformis, putrificans, proteus, etc.), which become pathogenic in the mortified tissues. Clinically, severe toxemia is observed as a result of toxic product absorption from the necrotic tissues. The liquefactive gangrene is encouraged by venous stasis.
In those cases where the affected tissues are contaminated by anaerobic bacteria (Clostridium perfringens, oedematiens, histolyticum, septicum, etc.), anaerobe or gas gangrene develops. The affected region gains an emphysematous aspect, which is bubbly at palpation due to the gas infiltration. It has a greenish-gray color and a putrefied smell (fig. 57).
The process extends extremely fast to the neighbouring tissues along the muscles, connective sheaths, vessels and other tissues leading to their necrosis. The respective microorganisms elaborate exotoxins which lead to severe intoxication and to the extension of the necrotic process. It represents a complication of the extended, open wounds produced in conditions of war, and road and work accidents which produce massive muscle and bone destruction. It is considered to be an independent infectious disease (primary gangrene).

- **Macrospecimen “Gangrene of the small intestine”** (fig. 58).

The intestine wall is edematous and black, with an opaque serous tunica. It is covered with fibrin deposits.

It is usually seen in atherosclerosis of the aorta and of the mesenteric artery, thrombosis of the mesenteric veins, and the strangling of the intestine in a hernial sac. It can be complicated with diffuse peritonitis, perforation of the intestinal wall, etc.

Another variety of necrosis is the eschar or decubitus necrosis. It represents necrotic foci of a blue-black color in the soft superficial tissues, the skin being, in many cases, ulcerated (fig. 59). It appears primarily in severe, cachectic patients with circulatory and neurotrophic disorders. It is severe in the regions that are exposed to long local mechanical compression (above the prominent parts of the bones) in sacral, trochanteric, scapular, calcanean regions, etc. It is observed especially in long-time immobilized bed patients in the same position (with malignant tumors, severe infectious diseases, cardiac insufficiency, etc.). The eschars gradually ulcerate, reaching the prominent parts of the bones.

5. **Fat necrosis**

Refers to the enzyme-mediated necrosis of adipose tissue, characterized by cleavage of neutral fat by lipase to triglycerides and fatty acids; the released fatty acids combine calcium and form insoluble salts that precipitate in necrotic foci. This necrosis result from liberation of powerful digestive enzymes into the substance of the pancreas and peritoneal cavity. It occurs in the acute pancreatitis and may be seen after trauma to fat.

6. **Fibrinoid necrosis**

Is manifested by the destruction of connective tissue (ground substance and collagen fibers) of the organ’s stroma and of the vascular walls; the necrotic masses infiltrate with plasma proteins (fibrinogen). The fibrinoid necrosis is characteristic for the imuno-allergic diseases (rheumatism, systemic lupus erythematosus, rheumatoid arthritis) and malign hypertension.

- **The consequences of necrosis** (table 5).

The substitution of the necrotic masses with connective tissue - organization (cicatrization) is one of the most frequent consequences of the necrosis of different organs. Other consequences are: a) encapsulation, calcification (petrification), ossification, sequestration – mostly seen in dry necrosis; b) the formation of cysts (cystic transformation), purulent lysis – in liquefactive necrosis; c) autoamputation and mummification are frequent consequences of the dry gangrene.
### The consequences of necrosis

<table>
<thead>
<tr>
<th>Description</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) Organization (cicatriztion)</td>
<td>The substitution of the necrotic site with connective tissue</td>
</tr>
<tr>
<td>b) Encapsulation</td>
<td>The formation of a membrane (capsule) of connective tissue around the necrotic site</td>
</tr>
<tr>
<td>c) Calcification (petrification)</td>
<td>The storing of insoluble calcium salts in the necrotic masses</td>
</tr>
<tr>
<td>d) Ossification</td>
<td>The substitution of the necrotic site with new formed bone tissue</td>
</tr>
<tr>
<td>e) Cyst formation (cystic transformation)</td>
<td>The appearance of some cavities after the lesion and reabsorption of the mortified tissue in liquefactive necrosis (more frequently in the brain and the spinal cord)</td>
</tr>
<tr>
<td>f) Sequestration</td>
<td>The detachment of the mortified tissue from the healthy tissue</td>
</tr>
<tr>
<td>g) Autoamputation</td>
<td>The total detachment from the body of some members or organs</td>
</tr>
<tr>
<td>h) Mummification</td>
<td>The drying of the mortified tissue in gangrene</td>
</tr>
<tr>
<td>i) Purulent lysis</td>
<td>The desintegration of the necrotic masses under the action of polymorphonucleated leukocytes in case of pyogenic overinfection</td>
</tr>
</tbody>
</table>

**APOPTOSIS**

Apoptosis (literally “falling off”) is a specialized, morphologically distinctive form of cell death, which should be differentiated from the common coagulative necrosis. It is a programmed and energy-dependent process of cell death designed to switch off and eliminate them (so-called cell suicide). Apoptosis occurs both physiologically and pathologically.

The elimination of cells by apoptosis is the main mechanism of cell death in several important physiologic processes and diseases, for example:
- removal of excess cells during embryonic development, e.g. limb development;
- elimination of cells in developmental involution, e.g. involution of thymus;
- elimination of cells in hormone-dependent physiologic involution (e.g. involution of lactating mammary gland epithelium after weaning and cyclic changes in endometrium during the menstrual cycle);
- clonal selection of lymphocytes in the induction of self-tolerance in development (deletion of autoreactive T cells in the thymus);
- elimination of cells in tissues that require a high cell turnover (intestinal lining epithelial cells);
- elimination of cells with acquired DNA damage through viral infection, ultraviolet or ionizing irradiation, cytotoxic agents (drugs);
- elimination of neoplastic cells in tumors;
- killing of viral infected cells by cytotoxic T-cells (e.g. viral hepatitis);
- death of nerve cells in neurodegenerative diseases (Alzheimer's disease).
Apoptosis usually involves single cell or clusters of cells. Initially the apoptotic cell lose surface specializations and junctions, shrinking in size, the nuclear chromatin condenses beneath the nuclear membrane. After that there is spitting of the cell into several fragments – apoptotic bodies. Apoptotic body is recognized in tissue sections as rounded or oval masses of intensely eosinophilic cytoplasm, containing some dense nuclear chromatin fragments and viable mitochondria and intact organelles. In final phase apoptotic bodies is recognized and phagocitosed by adjacent cells for destruction (fig. 60). The process takes a few minutes only, therefore this type of cell death is often hard to observe in vivo. This phagocytosis is clearly different from that seen in inflammation, when activated macrophages are recruited from outside the immediate area of death.

Apoptotic death can be triggered by a wide variety of stimuli, and not all cells necessarily will die in response to the same stimulus. Among the more important death stimuli is DNA damage (by irradiation or drugs used for cancer chemotherapy and by viruses).

Pathologic inhibition of apoptosis is observed in many neoplastic diseases and in several viral diseases.