Materials
for students for practical training in pathological anatomy
at the Department of Pathological Anatomy

II year Faculty of Dentistry

Topic: “Necrosis. Apoptosis ".

1. **The purpose of the lesson.** To study the issues of etiology, pathogenesis, morphology, complications and outcomes of irreversible cell damage (necrosis and apoptosis).

2. **Requirements for the level of the student for mastering the discipline - pathological anatomy. The student should know:**

1. Determination of damage to cells and tissues (reversible and irreversible).

2. Pathogenesis of irreversible damage.

3. Pathogenesis of ischemic injury.

4. Definition, etiology, pathogenesis, stages of necrosis, morphological manifestations and outcomes.

5. Definition, etiology, pathogenesis, stages of apoptosis, morphological manifestations and role in pathology.

Theoretical aspects.

Damage (injury) and death of cells and tissues

There are two main types of cell death - necrosis and apoptosis. Necrosis is the most common type of cell death due to exogenous influences, including ischemia and chemical stimuli. It is manifested by a sharp swelling or destruction of the cell, denaturation and coagulation of cytoplasmic proteins, destruction of cellular organelles.

Apoptosis - natural cell death - serves for the elimination (elimination) of unnecessary cell populations during embryogenesis and during various physiological processes. The main morphological feature of apoptosis is the condensation and fragmentation of chromatin. Although the mechanisms of necrosis and apoptosis are different, there are many similarities between these processes.

There are three main ***types of cell damage***: 1) ischemic and hypoxic; 2) caused by free oxygen radicals; 3) toxic.

In classical morphology, non-lethal cell damage is called ***dystrophy***, or reversible cell damage. Light-optical distinguishes between two types of such changes: swelling and fatty changes. Swelling develops when cells are unable to maintain ionic and fluid homeostasis. Fatal changes can also be a sign of reversible cell damage. Light-optically, they are characterized by the appearance of small or large lipid inclusions in the cytoplasm. They are found in hypoxic and various forms of toxic damage, mainly in cells involved in or dependent on fat metabolism, such as hepatocytes and myocardiocytes.

Necrosis is a spectrum of morphological changes that develop following cell death in living tissue. This is the result of the destructive effect of enzymes on a lethally damaged cell. In fact, two competing processes are developing: enzymatic cell digestion and protein denaturation.

Etiology of necrosis. Guided by the etiological factor, there are five types of necrosis: traumatic, toxic, trophoneurotic, allergic and vascular. Etiological factors can have a direct effect on tissue or indirectly - through the vascular, nervous and immune systems.

According to the mechanism of action of the etiological factor, necrosis can be direct and indirect. Direct necrosis can be traumatic, toxic. Indirect necrosis - trophoneurotic, allergic and vascular.

Traumatic necrosis is the result of a direct effect on the tissue of physical (mechanical, temperature, vibration, rational, etc.), chemical (acids, alkali, etc.) factors.

Toxic necrosis develops when tissues are exposed to toxic factors of bacterial and other nature.

Trophoneurotic necrosis is caused by impaired circulation and innervation of tissues in diseases of the central and peripheral nervous system. An example of trophoneurotic necrosis is pressure sores.

Allergic necrosis is the result of immune cytolysis of tissues during immediate or delayed hypersensitivity reactions. A classic example of allergic necrosis in immediate-type reactions involving immune complexes containing complement is fibrinoid necrosis in the Arthus phenomenon. Immune cytolysis with the participation of killer T-lymphocytes and macrophages leads to the development of liver tissue necrosis in chronic active hepatitis.

Vascular necrosis is associated with absolute or relative lack of circulation in the arteries, veins and lymphatic vessels. The most common form of vascular necrosis is caused by impaired blood circulation in the arteries due to their thrombosis, embolism, prolonged spasm, as well as functional overstrain of the organ under conditions of hypoxia. Insufficient circulation in the tissue causes their ischemia, hypoxia and the development of ischemic necrosis, the pathogenesis of which is associated not only with hypoxic, but also with reperfusion mechanisms.

Pathogenesis of necrosis. The mechanisms of necrosis are diverse, largely dependent on its etiology and structural and functional characteristics of cells, tissues and organs in which it develops. The end result of all pathogenetic mechanisms of necrosis is the emergence of intracellular chaos. Of the whole variety of ***pathogenetic pathways of necrosis***, five of the most significant can probably be distinguished:

- binding of cellular proteins with ubiquinone;

- ATP deficiency;

- generation of reactive oxygen species (ROS);

- violation of calcium homeostasis;

- loss of selective permeability by cell membranes.

Macroscopic signs of necrosis can manifest themselves in different ways: they depend on the originality of the organ in which necrosis occurs, as well as on the nature of the damaging factor. ***Microscopic signs*** relate to both the nucleus and the cytoplasm of cells, as well as the extracellular matrix.

Nuclea changes include:

- karyopyknosis - shrinking of the nuclei due to the condensation of chromatin;

- karyorexis - disintegration of nuclei into lumps;

- karyolysis - dissolution of the nucleus due to the activation of hydrolases (ribonuclease and deoxyribonuclease; RNase and DNase).

Changes in the cytoplasm are expressed in:

- plasma coagulation - protein denaturation and coagulation with the appearance of bright pink lumps in the cytoplasm;

- plasmorexis - the decay of the cytoplasm into lumps;

- plasmolysis - melting of the cytoplasm.

Changes in the extracellular matrix are manifested:

- in the splitting of reticular, collagen and elastic fibers under the influence of proteases, elastases, collagenases.

Necrotic masses are often impregnated with fibrin with the development of fibrinoid necrosis.

There are types of necrosis: coagulation, colliquation (wet), gangrenous (gangrene), caseous (cheesy, cheese-like) and fatty (enzymatic, or steatonecrosis), heart attack, sevestre.

Coagulative necrosis is the preservation of the general contours of the focus for at least several days. In this type of necrosis, damage or subsequently increasing intracellular acidosis denature not only structural proteins, but also enzymes and thereby block proteolysis of the cell. Coagulation necrosis is characteristic of hypoxic tissue death in all organs except the brain.

One of the most common types of such necrosis is a heart attack.

Infarction is tissue necrosis that occurs when blood circulation is impaired (vascular, ischemic necrosis). A heart attack develops as a result of thrombosis, embolism, prolonged spasm of the

arteries or functional overstrain of the organ in conditions of insufficient blood supply (the latter applies only to myocardial infarction).

Colliquation (wet) necrosis occurs as a result of autolysis or heterolysis, is more common in lesions with bacterial infectious agents and is due to the diluting action of proteolytic leukocyte enzymes. As for wet necrosis during hypoxic death of brain tissue, the reasons for its appearance remain unclear. The nature of such necrosis is explained by the fact that the brain tissue is rich in water and the processes of autolysis in it prevail over coagulation changes.

Gangrene is a black or dark brown necrosis that develops in the tissues, directly or through the anatomical channels in contact with the external environment. In addition to limbs, gangrene occurs in the lungs, intestines, cheeks, and elsewhere. The dark color of gangrenous tissue is caused by iron sulfite, which is formed from iron, hemoglobin and hydrogen sulfide in the air.

There are three morphological types of gangrene: dry, wet, and bedridden.

Dry gangrene is accompanied by mummification, a well-pronounced zone of demarcation inflammation, often occurs in the lower extremities. Macroscopically necrotic tissues (more often foot tissues) are reduced in volume, dry, black, and the demarcation zone is well defined.

Wet gangrene develops when dead tissue is infected with bacteria, usually anaerobic (for example, from the Clostridia group). With wet gangrene, the tissue swells, becomes edematous, the demarcation zone is not defined. This gangrene occurs in the intestines, lungs, uterus, limbs.

A bedsore is a type of dry or wet gangrene that occurs as a result of trophoneurotic disorders in weakened bedridden patients in areas of the body that are exposed to the greatest pressure.

Gas gangrene is rare and is an example of an infectious disease. With it, bubbles with hydrogen sulfide, usually produced by the microbe Clostridium welchii, are inside the necrotic tissue.

Caseous (cheesy, cheese-like) necrosis as a kind of coagulation is most often found in tuberculous foci. Macroscopically, it really resembles cottage cheese or soft cheese. Microscopically represented by structureless pink masses surrounded by a zone of granulomatous inflammation, consisting of tuberculous tubercles.

Fatty (enzymatic) necrosis, or steatonecrosis, is a focus of destroyed fatty tissue of a pale yellow color, putty-like appearance, of various shapes and sizes. More often this is a consequence of the release of activated pancreatic lipases, acting directly in the abdominal cavity in acute pancreatitis.

Sequestration is an area of dead tissue that does not undergo autolysis, is not replaced by connective tissue and is freely located among living tissues. Sequesters usually cause the development of purulent inflammation and can be removed through the resulting fistulous passages. Bone tissue undergoes sequestration more often, but sequestration can rarely be found in soft tissues.

The outcomes of necrosis are associated with reactive changes: processes of demarcation and repair, spreading from the zone of demarcation inflammation:

- organization, or scarring, - replacement of necrotic masses with connective tissue;

- encapsulation - delimitation of the area of necrosis by a connective tissue capsule;

- petrification - impregnation of the necrosis area with calcium salts (dystrophic calcification);

- ossification - the appearance in the area of bone tissue necrosis (occurs very rarely, in particular, in the foci of Gon - healed foci of primary tuberculosis);

- the formation of a cyst in the outcome of colliquation necrosis.

Under unfavorable circumstances, purulent fusion of necrotic masses occurs, and sepsis may develop.

Apoptosis. If necrosis is considered a pathological form of cell death arising as a result of a sharp damaging effect on the cell, then anontosis is opposed to it as a controlled process of cell self-destruction. The morphological manifestations of anontosis are the condensation of nuclear heterochromatin and cell shrinkage while maintaining the integrity of the organelles and the cell membrane.

The cell breaks down into apoptotic bodies, which are membrane structures with organelles and nuclear particles enclosed inside. Then the apoptotic bodies are phagocytosed and destroyed by lysosomes by the surrounding cells.

The characteristic signs of apoptosis are determined by the type of exposure and the type of cells. Chromatin condensation is associated with the cleavage of nuclear DNA, which occurs in the regions of bonds between nucleosomes and leads to the formation of fragments. Violation of the volume and size of cells is explained by the activity of transglutaminase. This enzyme catalyzes the cross-linking of cytoplasmic proteins that form a membrane under the plasma membrane. Phagocytosis of anontic bodies by macrophages and other types of cells is provided by the latter's receptors. One of the important features of apoptosis is its dependence on gene activation and protein synthesis. Induction of apoptosis-specific genes is provided by special stimuli such as heat shock proteins and protooncogenes. Several genes involved in the emergence and growth of malignant tumors (oncogenes and suppressor genes) play a regulatory role in the induction of apoptosis. For example, the p53 oncogene stimulates normal apoptosis.

Apoptosis is responsible for numerous physiological and pathological processes in the body:

- mediates the programmed removal of cells during embryogenesis (including implantation, organogenesis and involution);

- due to apoptosis, hormone-dependent involution of cells occurs in adults (for example, the rejection of endometrial cells during the menstrual cycle, atresia of follicles in the ovaries during menopause, regression of the lactating mammary gland after cessation of feeding the child);

- ensures the destruction of cells in proliferating cell populations, such as the crypt epithelium of the small intestine, and cell death in tumors;

- death of autoreactive clones of T-lymphocytes and pathological atrophy of hormone-dependent tissues (for example, atrophy of the prostate gland after castration and the disappearance of lymphocytes in the thymus after the injection of glycoproteins) are realized through apoptosis;

- apoptosis underlies the pathological atrophy of the parenchymal organs after the closure of the duct (for example, the pancreas, parotid salivary gland, kidney);

- apoptosis is associated with cell death caused by cytotoxic T cells (for example, in the case of transplant rejection), and cell death in certain viral diseases (for example, in viral hepatitis, in which cell fragments during apoptosis are known as Kaunsilman's bodies) ;

- apoptosis underlies cell death caused by various weak damaging effects, which in large doses lead to cell death (thermal effects, radiation, cytotoxic anticancer drugs and, possibly, hypoxia).

Infarction. This is tissue necrosis due to the cessation and significant decrease in arterial blood flow. However, the likelihood of a heart attack in various organs is not the same, since the degree of development of collateral circulation is of great importance.

In some organs, such as, for example, in the brain, there are anastomoses that provide collateral blood supply when one of the arteries is blocked or clamped, while a cerebral infarction may not develop. Thus, the lungs and the liver are relatively resistant to ischemia due to the fact that when the branches of the pulmonary or hepatic artery are occluded, the deficiency of blood filling is compensated by double blood supply from the systems of the bronchial arteries and the portal vein.

If the turning off of the main artery occurs slowly, then the restoration of blood supply is also gradual, therefore, even with a blockage of the main artery, a heart attack may not develop immediately.

Interruption of venous drainage, which slows down and even stops arterial inflow, can also lead to a heart attack, but most often a heart attack occurs when a blood clot forms or a vessel is blocked by an embolus.

Macroscopically, heart attacks are divided into white (anemic) and red (hemorrhagic).

White infarctions are found in the myocardium, spleen, kidneys, brain; are represented by a grayish-white structureless tissue that acquires a yellowish tint 48-64 hours after its development. Between the focus of the infarction and the unchanged tissue in the myocardium and kidneys, a zone of hyperemia in the form of a hemorrhagic corolla is clearly visible. In the spleen, the hemorrhagic corolla is not visible, since it merges with the crimson background of the pulp. White infarctions are most often dry (coagulative), and wet (colliquation) necrosis in the brain tissue.

Red infarctions occur when arteries and veins are occluded in the lungs, small intestine, ovaries, and sometimes in the brain. In the development of hemorrhagic infarctions, the presence of venous stasis, as well as a double type of blood supply, are of great importance. As a rule, the occurrence of red heart attacks is also associated with blockage of arteries by blood clots or thromboembolism (for example, branches of the pulmonary artery). However, with dynamic

torsion of intestinal loops and their infringement in the hernial sac, vein constriction appears, which leads to hemorrhagic intestinal infarction.

The shape of a infarction depends on the angioarchitectonics of the organ, the severity of collateral blood supply and can be wedge-shaped (triangular) and irregular. In the first hours of development, macroscopically, the infarction can be difficult to distinguish, and only pallor or hemorrhage allows it to be diagnosed. If the infarction reaches the capsule or serous membrane of the organ, then fibrin often appears on them.

Final diagnosis and determination of the time of development of a heart attack are possible with microscopic examination. On the periphery of the focus of necrosis, edema, hyperemia develop; leukocytes, macrophages and mast cells accumulate, a zone of demarcation (i.e., delimiting) inflammation is formed. The accumulation of leukocytes containing proteolytic enzymes, and partial resorption contribute to softening, dissolution and can lead to rupture of necrotic tissue. For example, as a result of rupture of a necrotic muscle and hemorrhage in the pericardial cavity, cardiac arrest occurs. Along with infiltration, the number of newly formed vessels increases and granulation tissue is formed, the maturation of which leads to the formation of a scar at the site of a heart attack. This process is called organization.

In the kidneys in the area of the scar, retractions are formed, and the surface of the kidney becomes uneven, coarse. In the brain, with small sizes of the necrosis zone, a small glial scar can form, with large ones - a cavity filled with fluid (kit). Hemorrhagic heart attacks of the lung with the addition of an infection and the development of purulent inflammation can melt. Hemorrhagic intestinal infarctions are usually complicated by the occurrence of intestinal gangrene, wall perforation, and fecal peritonitis.

3. Lesson plan

Macropreparations

1. **Ischemic cerebral infarction** - pay attention to the shape, consistency and color of the necrosis focus.

2. **Ischemic spleen infarction** - pay attention to the shape, color and consistency of the necrosis focus.

3**. Gangrene of the foot** - pay attention to the volume of necrotic tissues, their color and consistency, note the presence of a demarcation line.

4. **Gangrene of the intestine** - note the state of the mucous membrane, color, thickness, consistency of the intestinal wall, the state of the serous membrane and mesenteric vessels.

5. **Tuberculosis of the lymph nodes** - pay attention to the color, shape, consistency of foci of necrosis in the lymph nodes.

6. **Pancreatic necrosis** - pay attention to the color, shape, texture and localization of necrotic changes.

7. **Ischemic myocardial infarction with a hemorrhagic corolla** - pay attention to the shape, color, consistency of the necrotic area, changes in the pericardium in the infarction zone.

8. **Hemorrhagic pulmonary infarction** - pay attention to the shape, color, consistency of the necrotic area, changes in the pleura in the infarction zone.

Micropreparations

1. **Apoptotic bodies (Kaunsilman's little bodies) in hepatitis** (staining with hematoxylin and eosin) - pay attention to the localization, shape, structure and color of Kaunsilmen's little bodies.

2. **Necrosis of the epithelium of the convoluted tubules of the kidney** (staining with hematoxylin and eosin) - pay attention to the state of the nuclei and cytoplasm of the epithelium of the tubules, the blood filling of the capillaries of the glomeruli and vessels of the medulla of the kidney.

3**. Ischemic renal infarction** (staining with hematoxylin and eosin) - pay attention to changes in the necrosis focus and the zone of demarcation inflammation.

4. **Myocardial infarction** (staining with hematoxylin and eosin) - pay attention to changes in the zones of necrosis, demarcation inflammation and intact tissue.

5. **Hemorrhagic pulmonary infarction** (staining with hematoxylin and eosin) - pay attention to changes in the areas of necrosis, demarcation inflammation and intact tissue.

Electronograms

1. **Apoptotic body** - pay attention to changes in chromatin, the structure of the apoptotic body.

Situation cases.

Situation case 1

Patient A., 75 years old, suffering from atherosclerosis, was taken by ambulance to the city clinical hospital with a clinical picture of an acute abdomen. During the examination, intestinal obstruction was diagnosed. The patient was operated on. When the abdominal cavity is opened, the loops of the small intestine are swollen, black in color, the mesenteric vessels are obturated with thrombotic masses.

Questions to the situation case 1

1. What process has developed in the patient in the small intestine that caused small intestinal obstruction?

2. The cause and pathogenesis of this process in the intestine?

3. Explain the origin of black intestine?

4. Describe the macroscopic preparation of the small intestine.

5. With what pathological processes should this pathological process be differentiated?

6. In what tissues does this pathological process develop?

7. List the main types of necrosis.

Situation case 2

Patient B., 23 years old, with a gunshot wound to the neck died from acute renal insufficiency as a result of massive blood loss and developed posthemorrhagic shock. An autopsy revealed anemia of internal organs.

Questions to the situation case 2

1. What pathological process has developed in the kidneys?

2. Name the type of this pathological process according to its etiology.

3. What is the pathogenesis of the pathological process in the kidneys?

4. Describe the micropreparation that demonstrates the pathological process in the kidneys.

5. Describe the condition of the tubular basement membranes.

6. Name the favorable outcome of this pathological process in the kidneys.

7. Name other etiological factors that can cause a similar pathological process in the kidneys.

Situation case 3

Patient V., who died from an ischemic cerebral infarction, has a pressure ulcer in the sacrum area on the section.

Questions for situation case 3

1. What pathological process has developed in the brain tissue, its

names depending on consistency and color?

2. Name the type of this pathological process by etiotropic classification and its causes.

3. What is the pathogenesis of the pathological process in the brain tissue?

4. Describe a macro specimen demonstrating a pathological process in the brain tissue.

5. Name the favorable outcome of this pathological process in the brain tissue.

6. Name the pathological process that has developed in the region of the sacrum.

7. List the most significant mechanisms in the pathogenesis of irreversible tissue damage.

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