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## Identification of time of death of cats according to histological changes in some organs

J. Serdioucov\*, D. Shkundia\*, O. Kruchynenko\*\*

\*National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine

\*\*Poltava State Agrarian University, Poltava, Ukraine

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National University of Life  
and Environmental Sciences  
of Ukraine, Heroiv  
Oborony st., 15/3,  
Kyiv, 03041, Ukraine.

Poltava State Agrarian  
University, Skovorody st., 1/3,  
Poltava, 36003, Ukraine.  
Tel. +38-099-062-64-96.  
E-mail:  
oleg.kruchynenko@pda.u.edu.ua

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Determining the time of an animal's death is an extremely relevant subject in the practice of modern forensic medicine, because the issue often arises when investigating crimes related to animal abuse, in particular, with domestic cats *Felis silvestris catus* (Linnaeus, 1758). Methods of identifying time of cats' death are currently developed insufficiently and their introduction into the veterinary-forensic practice has been limited, including the histological method we have employed. We used five corpses of cats, from which we periodically, after certain time periods, gathered the material for histological study, sampling such organs as the liver, kidneys, lungs, heart (myocardium), and the skeletal muscles. From the collected material, we made histological sections and stained them with hematoxylin and eosin. We determined the general patterns in changes occurring in the internal organs of cats over time, namely changes in the staining intensity of the cell elements and intercellular structures on the histopreparations; formation of detritus-filled cavities; a number of distinctive changes in the connective-tissue stroma of the organs; emergence of bacilliform bacteria in the tissues and formation of their colonies. In the muscle tissue, we found specific features such as loss of alignment and emergence of transversal grooves with subsequent fragmentation. The study revealed that the development of those changes in each organ clearly correlated with time that has passed since each animal had died. Thus, a complex evaluation of detected changes can give a more accurate – compared with other existing methods – assessment of the time of death of a corpse submitted to forensic veterinary examination. We recommend using the histological method for animals presumed to have been dead for no more than 18 days, because later the tissue disintegration reaches such a level that complicates the correlation of changes with time. We consider it promising to study the time of death of domestic cats using other methods for identifying criteria for postmortem intervals longer than 18 days, and also identifying time of death in other species of animals.

**Keywords:** identifying time of death; *Felis silvestris catus*; microscope changes; internal organs; skeletal muscles.

### Introduction

As of now, animal abuse is a relevant problem in society. It is actions of some persons towards animals, leading to injuries, disabilities or death (Parry & Stoll, 2020; Rebollada-Merino et al., 2020). Often, objects of abuse are domestic cats – some of the commonest pets (Huang et al., 2020; Rebollada-Merino et al., 2020). Death of cats can result from injuries inflicted using weapons, both firearms and cold weapons, and also various objects or tools (Doukas, 2022). There have been episodes when the animals were thrown from height, asphyxiated in some way or another, tortured, or poisoned (Romano et al., 2020), leading to lethal outcomes (Nation, 2021). Animals can be found right after their death or at the moment of death, but oftentimes they are found after some time, and then the authorities investigating the case face the necessity of identifying the postmortem interval. Corpses of animals, in particular, domestic cats, are the commonest objects of expert examination in the sphere of veterinary forensic medicine. Identification of the date and time when an animal had died is often necessary for inquest and investigation; to confirm correspondence of testimonies of persons that represent the sides of a case and judicial process; for checking accuracy of the anamnesis prepared for the performance of pathoanatomical autopsy of an animal.

A large number of methods have been developed for identifying time of death (Hryhorian, 2019; Voronov, 2022). We should note that most of them have been designed for the sphere of forensic expertise of human corpses (Sarkisova, 2021; Savka et al., 2021; Davydenko, 2022; Lytyvnenko, 2022; Sokol, 2022; Voronov, 2022), and only a small proportion of them are used to examine animal corpses (Sapienza et al., 2020).

The classical methods are those most commonly used today, such as visual-palpatory, thermometry (Jeong et al., 2020); various biochemical methods, namely studying expression and concentration of proteins in the skeletal muscles (Saber et al., 2021; Piegari et al., 2023), concentrations of various substances in the vitreous body of corpse eyes (Garland et al., 2020; Perez-Martínez et al., 2020; Ave et al., 2021; De-Giorgio et al., 2021; Nioi et al., 2021; Rajamani et al., 2021; Sarkisova, 2021); methods of image analysis, such as computer tomography (Watson & Baucom, 2020; Yamada et al., 2023), magnetic resonance imaging (Sapienza et al., 2020; Yitbarek & Dagnaw, 2022; Zhang, 2022), and elastography (Garczynska et al., 2021). Other common methods are forensic-entomological examination (Byrd & Sutton, 2020; Byrd & Sutton, 2021; Ivorra et al., 2021; Matuszewski, 2021; Weidner & Hans, 2021; Ashraf, 2022), forensic taphonomy (Miles et al., 2020), DNA studies (Mori et al., 2021; Heba El-Sayed et al., 2023), and dental profiling (Viciano et al., 2022).

A separate mention should be made for identification of time of death according to the results of histological studies of the tissue samples from a corpse, used in both forensic medicine and forensic veterinary expertise (Ceciliason et al., 2021; Delgado et al., 2021; Saber et al., 2021; Sokol, 2022). The method consists in evaluation of autolysis- and decay-related (breakdown changes) changes in the tissues. The degree of development of those changes correlates with the time period that has passed since death (Mohammed et al., 2023). Besides the histological method, forensic medicine now also employs its more modern variations such as spectropolarimetry (Sarkisova & Malanchuk, 2020) and reconstruction of the polycrystalline structure of histopreparations (Lytyvnenko, 2022). In the sphere of forensic veterinary medicine, compared with forensic medicine, there

are only some fragmented studies focusing on identification of postmortem interval using the histological method. No peculiarities of the material taken from corpses of various animals have been reported, in particular of cats; animal breeds; sex and age groups, etc.

## Materials and methods

The research was carried out during 2021–2023 at the V. H. Kasiienko Department of Anatomy, Histology, and Pathomorphology of Animals of the National University of Life and Environmental Sciences of Ukraine (Kyiv). The material was collected from corpses of animals involved in the experiment, and its further analysis was performed according to the ethical norms of the international and Ukrainian legislation. In the experiment, we used 5 corpses of domestic cat (*Felis silvestris catus*), of which 2 were Maine Coon, 1 Burmese, and 2 non-pedigree. All the animals were females, aged 2–3 years, moderately fed. The department received the animal cadavers for the research from various veterinary clinics of Kyiv after they had been euthanized according to the recommendations of veterinary doctors and the owners' approval. We conducted a pathoanatomical necropsy of the animals' corpses, according to the generally accepted methods (without evisceration). For the microscop-

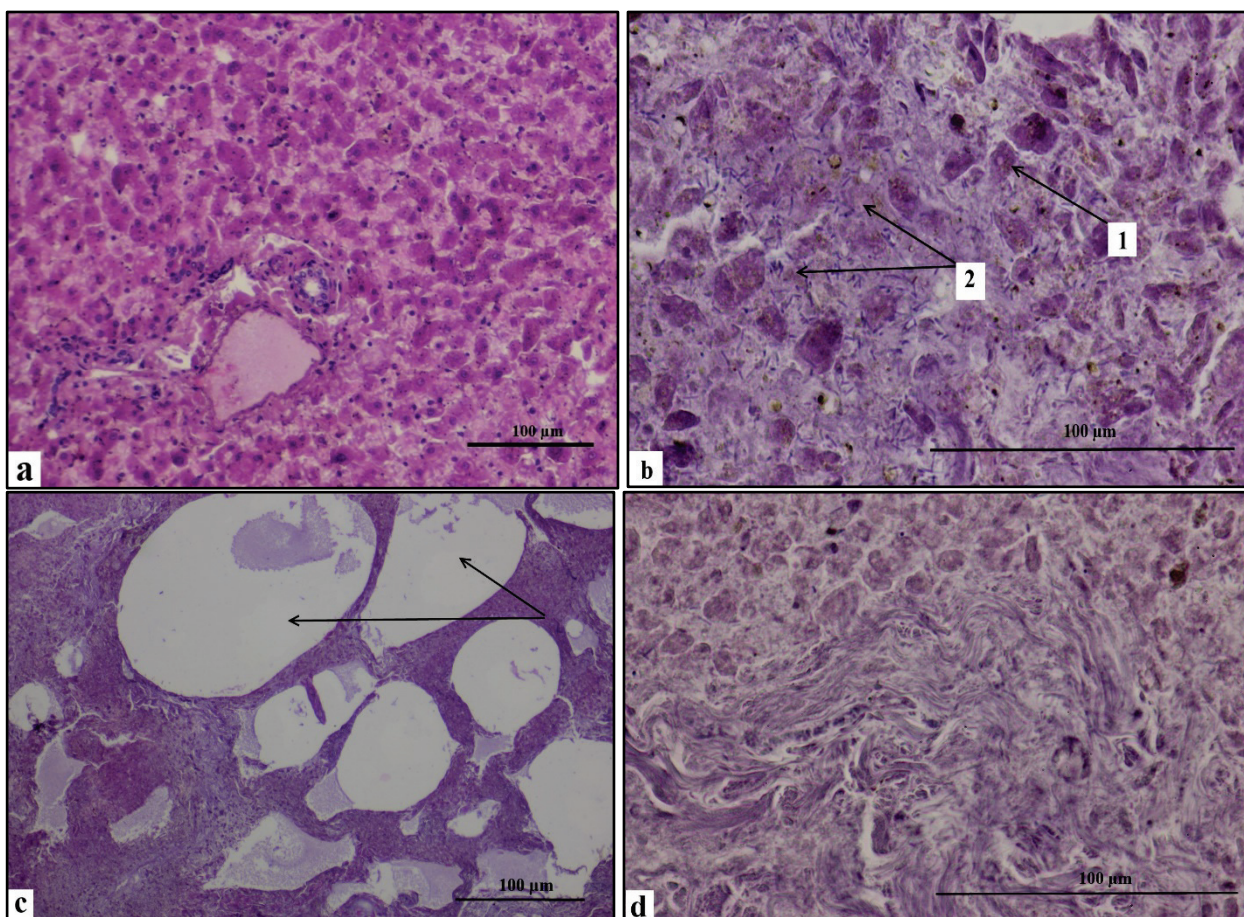
ic study, we sampled the liver, kidneys, lungs, myocardium, and skeletal muscles from the area of the thigh (if no visible pathoanatomical changes were present in those organs).

Then, the corpses of the experimental animals were stored in sterile conditions, at room temperature (20 °C). The same organs were repeatedly sampled for microscopic examination at 3-day intervals. The corpses were stored until the 18th day, inclusive, because at later periods the material became unsuitable for further use as a result of significant spread of decay.

The sampled material was fixated in 10% buffer formalin solution according to Lilly (1969); and submerged in paraffin. We prepared 5–10 µm-thick histosections, which were stained using Leica hematoxylin and eosin and examined under a light microscope msq 2000, preparing the necessary number of microphotographs.

## Results

The microscopic structure of the liver in the 0–3 days postmortem interval (Fig. 1a) corresponded to the norm of the histostructure of this organ. In the 4–6 day postmortem interval, we observed the liver to have pronounced disintegration of the hepatic laminae. Hepatocytes did not form clearly aligned linear structures, typical for the hepatic laminae.



**Fig. 1.** Cat liver: *a* – 0–3 days postmortem interval, general view; *b* – 4–6 days postmortem interval, vacuoles (cavities) in the place of hepatocyte nuclei (1) and bacterial cells inside the lobules (2); *c* – 7–9 days postmortem interval, vacuoles that contain detritus; *d* – 10–12 days postmortem interval, connective-tissue fibers of wavy configuration; hematoxylin and eosin

However, their residues were found in some places. Some areas of the hepatic lobules were ruined and appeared as structures of irregular shape, filled with homogenous pale-eosinophile substance with individual basophilic inclusions. In some regions, in the places of hepatic lobules, we saw round or oval cavities, sometimes divided into several parts by membranes. Most hepatocytes maintained their form, nuclei in them stained badly, but were visible under large increase in the magnification of the light microscope. Sometimes, in the place of lost nuclei, we detected vacuoles (cavities) (Fig. 1b). Cytoplasm of hepatocytes was granular, non-uniform in structure, and some areas of it were stained badly. In the con-

nective tissue of the liver, we saw ruination of collagen fibers and less intensive colour of nuclei of fibroblasts and fibrocytes. Large areas of the connective tissue were uniform in appearance. Configuration of the connective-tissue fibers was wavy in some regions. Ubiquitously, both in the connective tissue and inside the lobules, we found colonies of *Bacillus* bacteria between hepatocytes, stained basophilously (Fig. 1). In the connective tissue, colonies of bacteria in some areas were a network that had permeated the thickness of the tissue. Inside the hepatic lobules, between hepatocytes, accumulations of grains of pigment (bilirubin) were found. In the 7–9 days postmortem interval, the microscopic structure of the cats'

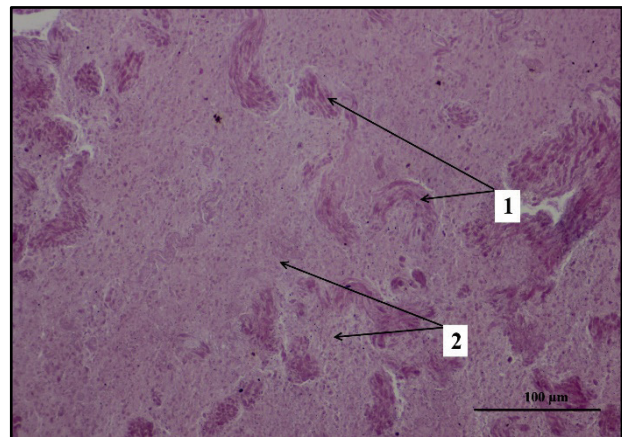
liver was similar to that in the previous time interval. Disintegration of hepatic lobules was pronounced completely; no aligned structures typical for the hepatic laminae in the norm were seen in the fields of view. Hepatocytes did not contact with each other, no nuclei were found, and some hepatocytes had cavities in places of nuclei. Cytoplasm of hepatocytes was non-homogenous, unevenly stained, and contained a large number of grains of pigment (bilirubin). Between hepatocytes, bacterial colonies or single bacterial cells were seen. In places of some lobules, we saw emergence of large vacuoles, partly filled with detritus, which contained a large amount of bacteria (Fig. 1c). The tissue of hepatic lobules and even the inter-lobular connective tissue contained a large amount of pigment (bilirubin). In some places, we observed fragmentation of the bundles of fibrous connective tissue. The fibers that maintained integrity were loose and often became of wavy configuration. In the 10–12 days postmortem interval, hepatocytes in each field of view had no nuclei. Cytoplasm of hepatocytes was non-homogenous, unevenly stained, and many hepatocytes had completely unstained areas of cytoplasm. Between hepatocytes, we observed a large amount of accumulations of detritus in the form of homogenous, grainy, non-structured substance, and accumulation of pigment (bilirubin). The connective tissue had wavy configuration in some places (Fig. 1d).

In the 13–15 days postmortem interval, the structure of the hepatic lobules was not observed. In fields of view, we saw some regions that contained hepatocytes – residual hepatic lobules (Fig. 5). Around them, there were large areas that were homogenous, non-structural, poorly eosinophilous substance – detritus (Fig. 2). Hepatocytes were poorly stained with eosin, having no structure typical for nuclei and cytoplasm. The connective tissue in some areas was completely ruined. Such regions had the appearance of unstained sections between the residuals of hepatic lobules, and in some areas, they had wavy form and signs of partial fragmentation. Inclusions of bilirubin were found between remaining elements of the liver tissue.

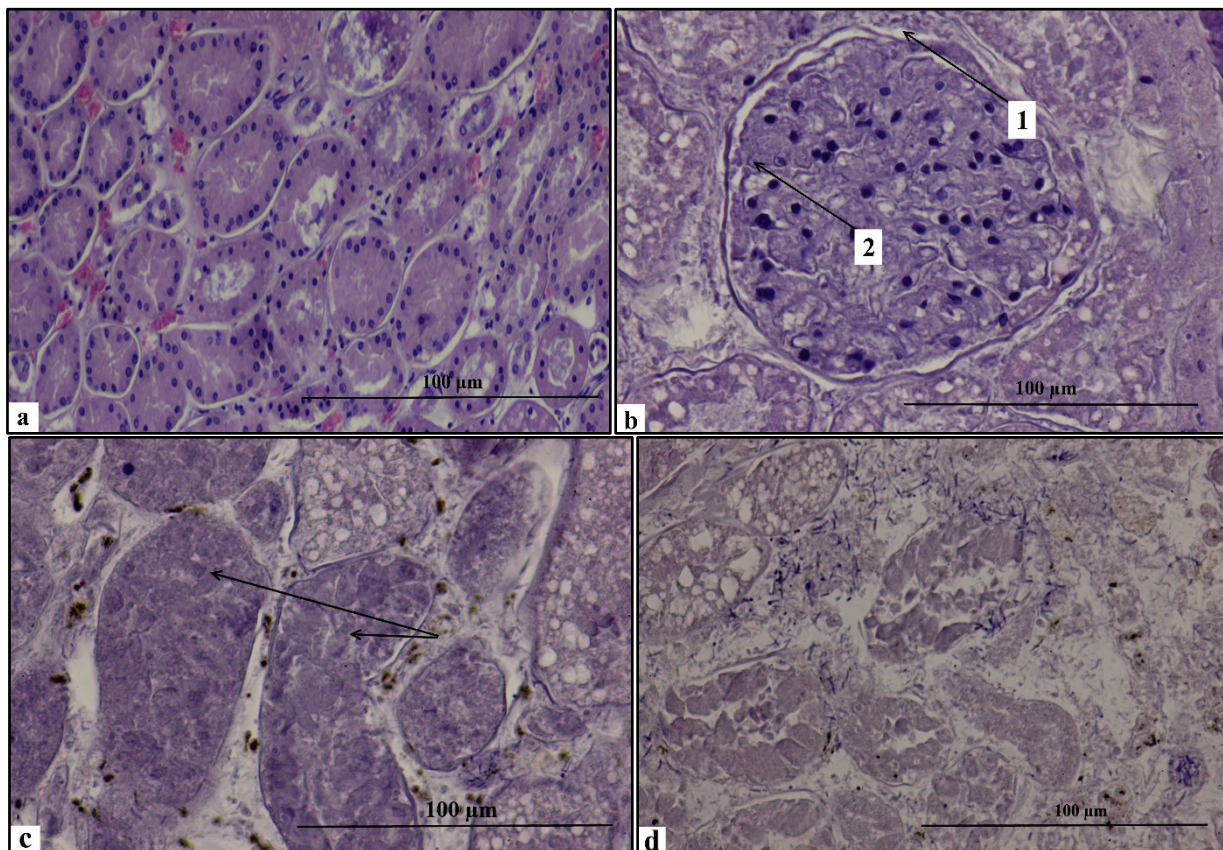
In the 16–18 days postmortem interval, we saw giant colonies of bacteria in places with residues of the connective tissue. Smaller colonies

were observed in the detritus substance. The number of hepatocytes looked significantly reduced.

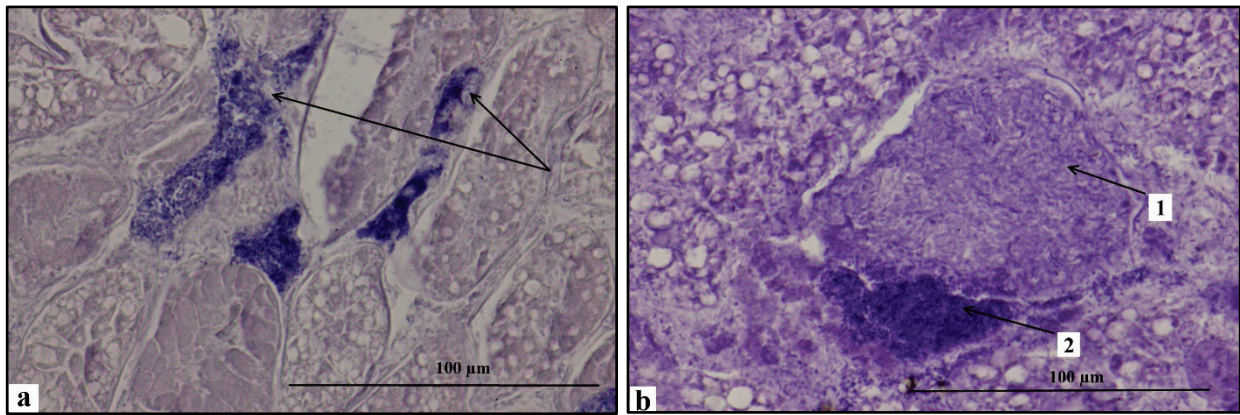
The microscopic structure of the kidneys in the 0–3 days postmortem interval corresponded to the histostructure of this organ in the norm (Fig. 3a). In the 4–6 day interval, we saw unstained sections between the vessels of bundle. The epithelium of the capsule of the renal corpuscle was detaching from some places and was partly in the lumens of the corpuscles (Fig. 3b). The corpuscle had a somewhat wavy configuration (Fig. 3b). Lumens of the renal canals were narrowed. The epithelial cells were larger, and some epitheliocytes had no nuclei at all, with distinctive cavities instead. In some regions, we found partial fragmentation of the cytoplasm of epitheliocytes, which was homogenous and appeared as turbid-grey. Collagen fibers of the intermediate connective tissue had partly loosened fibers and were of somewhat wavy configuration.



**Fig. 2.** Cat liver, 13–15 days postmortem interval: residuals of the hepatic lobules (1), detritus (2); hematoxylin and eosin



**Fig. 3.** Cat kidney: a – 0–3 day postmortem interval, general view; b – 4–6 day postmortem interval; wavy configuration of the capsule (1), detachment of capsule epithelium (2); c – 7–9 day postmortem interval, absence of lumens in the ducts; d – 10–12 day postmortem interval, bacterial cells (blue, rod-like) in the connective tissue; hematoxylin and eosin



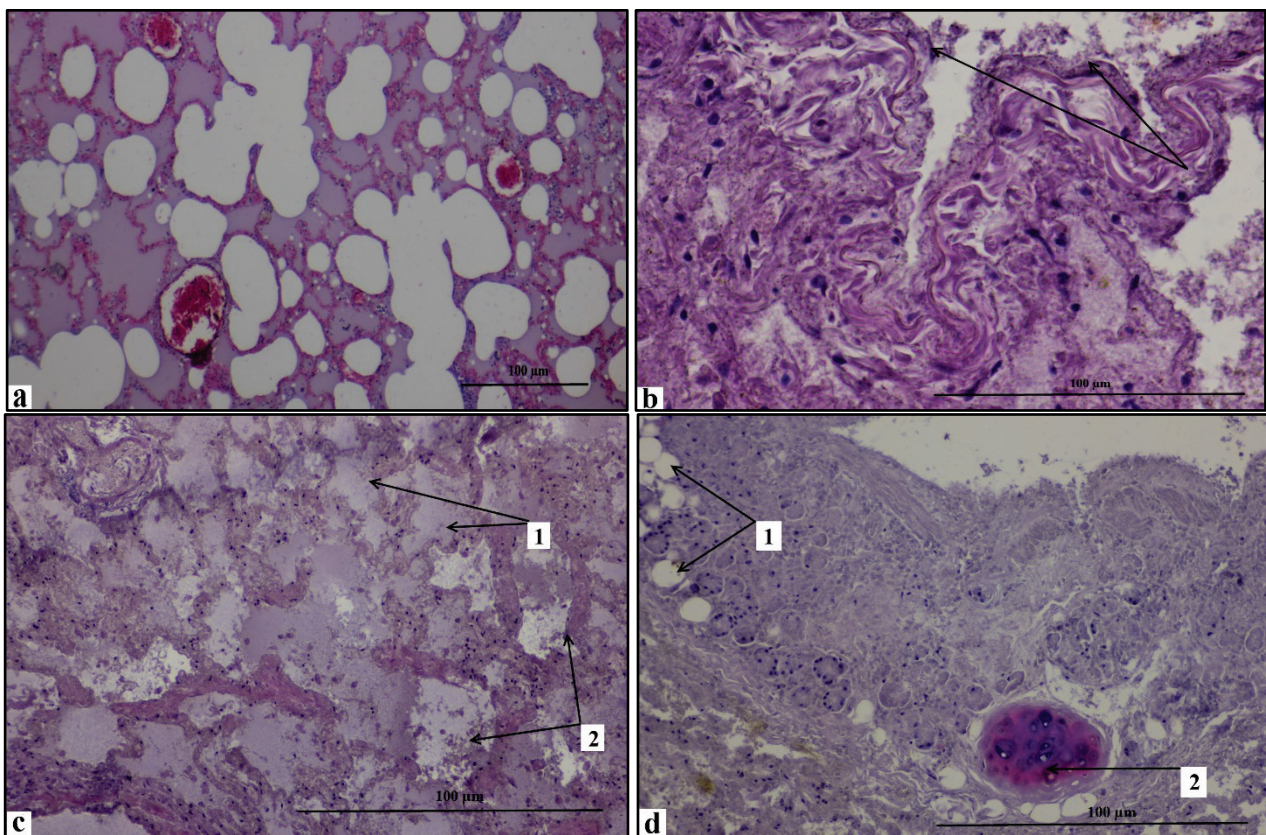
**Fig. 4.** Cat kidney: *a* – 13–15 day postmortem interval, bacterial colonies; *b* – 16–18 day postmortem interval, residuals of the renal corpuscle (1), colonies of bacteria (2); hematoxylin and eosin

In the 7–9 day interval, nuclei of podocytes, mesangial cells, and endothelial cells in the vessel bundles were partially stuck together. Epithelium that covers the capsule was completely ruined. In the lumen between the capsule and vascular bundle, non-homogenous, non-structured, and poorly eosinophilous substance was seen. In the canals, there were practically no lumens, and no nuclei of epithelial cells were detected (Fig. 3c). We saw pronounced fragmentation of cytoplasm of epithelial cells. The sections between the ducts and renal corpuscles visually looked enlarged. The intermediate connective tissue was stained poorly, the collagen fibers were loose, and its thickness was observed to have some rod-like bacterial cells, stained basophilically. In the time interval of 10–12 days, the changes in the renal corpuscles were similar to such in the previous time interval. All the structures of the kidneys were stained poorly. Also, we observed notable fragmentary breakdown of the ductal epithelial cells. In the intermediate connective tissue, and also some ducts, we observed formation of bacterial colonies, and also a large amount of single

bacterial cells (Fig. 3d). In some places, the intermediate connective tissue appeared as a granular, non-structural mass.

In the 13–15 day interval, the changes looked similar to those in the 10–12 day interval. We saw formation of large bacterial colonies (Fig. 4a), which were located mostly between the ducts and renal corpuscles. In the time interval of 16–18 days, the renal corpuscles had irregular shape and wavy contours. Cellular elements of the renal corpuscles had no nuclei, and vascular bundles appeared as homogenous, non-structural mass, in some places containing large colonies of bacteria (Fig. 4b). Most of renal ducts were ruined. In the intermediate connective tissue, bacterial colonies were large, occurring in large numbers.

The microscopic structure of the lungs in the 0–3 day postmortem interval corresponded to the histostructure of this organ in the norm (Fig. 5a). In the interval of 4–6 days, alveoli were of irregular shape. Their contours were wavy, and the lumens of the alveoli were often smaller than the normal.



**Fig. 5.** Cat lungs: *a* – 0–3 day postmortem interval, general appearance; *b* – 4–6 day postmortem interval, wavy configuration of the intermediate-connective-tissue fibers; *c* – 7–9 day postmortem interval, irregular shape of alveoli (1), detritus in the lumens of alveoli (2); *d* – 10–12 day postmortem interval, cavities in the bronchial wall (1), eosinophilous staining of the cartilage (2); hematoxylin and eosin

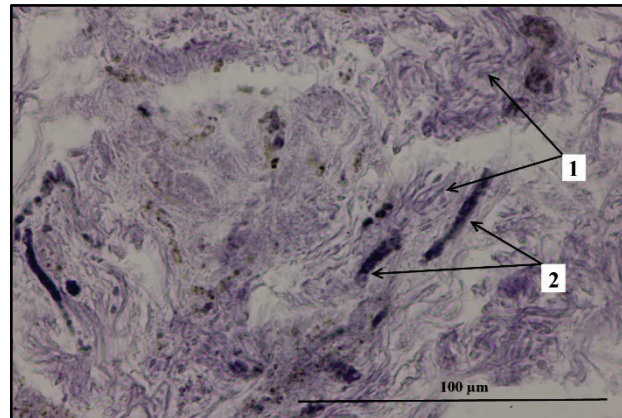
The lumens of the alveoli were often filled with swollen, non-homogenous, poorly-eosinophilous substance, in which we found single cells of the alveolar epithelium, separated from the basal membranes. The nuclei of alveolocytes were stained poorly. In the bronchi and bronchioles, we observed partial detachment of the mucous membrane and its fragments were in the lumens. In the intermediate connective tissue, configuration of collagen fibers became wavy (Fig. 5b). Cells of the intermediate connective tissue had apparent nuclei. In the blood vessels, the content was homogenous and eosinophilously stained.

In the 7–9 day interval, the alveoli were of irregular shape (Fig. 5c), epithelial layer of the alveolar wall was ruined, alveolar cells were found in the alveolar lumens. No nuclei were seen in them. In some places, the inter-alveoli membranes were ruined and neighbouring alveoli fused together. The mucous membrane of the bronchi and bronchioles was almost completely devoid of epithelium. The lumens of bronchi and alveoli (Fig. 5c) contained homogenous, non-structural, poorly stained substance (detritus). In the bronchial walls, we observed some round shape cavities. The cartilage tissue of the walls of average-diameter bronchi was stained eosinophilously. On some preparations, we observed partial ruination of the walls of the bronchi and bronchioles. In the intermediate connective tissue, the collagen fibers were loose and had a wavy configuration. In the content of the blood vessels was a granular and non-structure substance. In the connective tissue, especially the tissue surrounding vessels, and also in the lumens of some blood vessels, we saw single bacterial cells, stained basophilously.

In 10–12 days interval, the lumens of the alveoli were reduced, their contours were of irregular shape, and formed tortuous folds. In some places, the alveoli walls were ruined. In such places, there formed areas covered by non-structured, non-homogenous, poorly stained substance. The same substance was found in the lumens of alveoli. The mucous membrane of bronchi and bronchioles had almost no epithelium. In the lumens of bronchi, we saw homogenous, non-structured, poorly stained substance (detritus). In the bronchial walls, some rounded cavities were found (Fig. 5d). The cartilage tissue of the walls of average-diameter bronchi in some places was stained eosinophilously (Fig. 5d). On some preparations, we observed partial ruination of the walls of bronchi and

bronchioles. In the intermediate connective tissue, the collagen fibers were deformed. In the connective tissue, especially the one surrounding the blood vessels, and also in the lumens of some blood vessels, we saw single bacterial cells, stained basophilously.

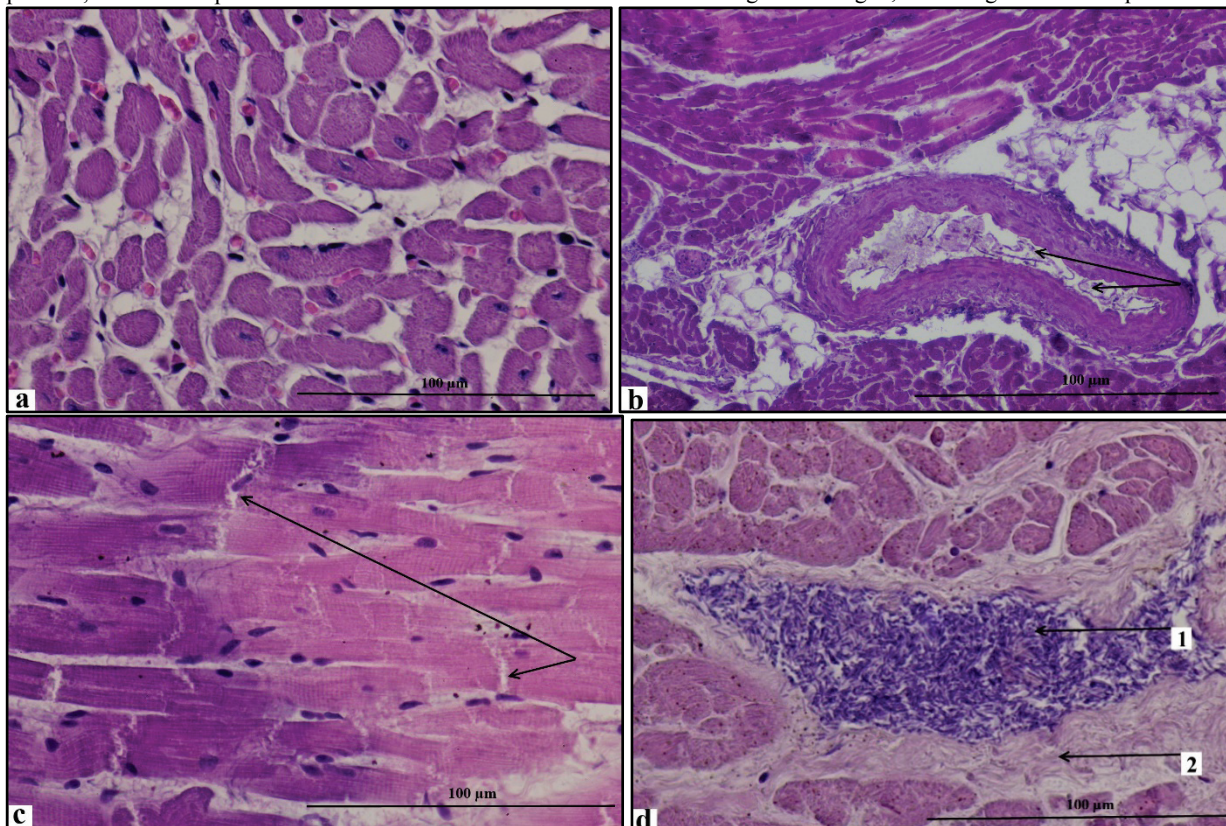
In 13–15 day interval, the lung tissues appeared as a homogenous, non-structured, poorly stained substance, the layer of which contained numerous large cavities. This mass had areas of expressed basophilous colour – colonies of bacteria (Fig. 6). The connective tissue constituted areas containing fragments of collagen fibers of wavy configuration (Fig. 6).



**Fig. 6.** Cat lungs, 13–15 day postmortem interval, fragments of connective-tissue fibers of wavy configuration (1), colonies of bacteria (2); hematoxylin and eosin

In the 16–18 day interval, the changes were similar to the previous time interval. In the blood vessels, we observed a non-homogenous, non-structured mass with bacterial cells.

The microscopic structure of myocardium in the 0–3 day postmortem interval corresponded to the histostructure of this organ in the norm (Fig. 7a). In the 4–6 day time interval, the nuclei of cardiomyocytes underwent staining to lower degree, and the alignment in some places remained.



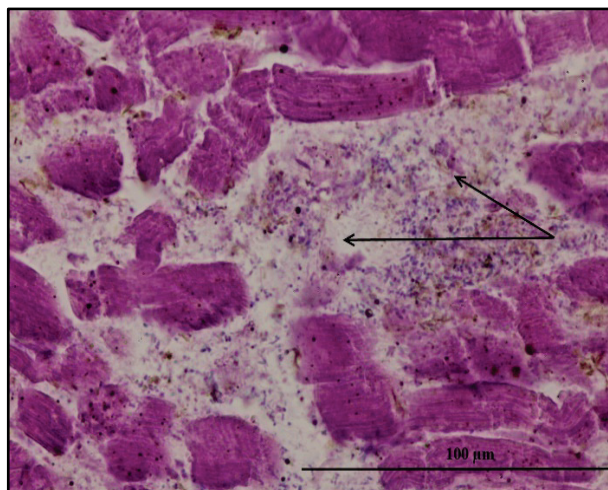
**Fig. 7.** Cat myocardium: a – 0–3 day postmortem interval, general view; b – 4–6 day postmortem interval, bacterial cells in the lumen of blood vessels; c – 4–6 day postmortem interval, transversal grooves in the muscle fibers; d – 7–9 days post-mortem interval; colony of bacteria in the intermuscular connective tissue (1), wavy configuration of the connective-tissue fibers (2); hematoxylin and eosin

In some places of the layer of muscle fibers, we saw transversal grooves (Fig. 7c). The intermuscular connective tissue was altered. In it, collagen fibers were loose and misaligned. We observed colonies of bacteria, of irregular shape, which were between the bundles of muscle fibers. Single bacterial cells were also found in the lumens of blood vessels (Fig. 7b). In the 7–9 day interval, no nuclei of cardiomyocytes were seen. Alignment partly remained. In the cytoplasm of cardiomyocytes, the transversal grooves were well manifested. In some regions, we observed fragmentary disintegration of the muscle fibers. In the muscular connective tissue, collagen fibers were misaligned, loose, sometimes of wavy configuration (Fig. 7d). A distinctive feature was formation of large bacterial colonies (Fig. 7d), located between the bundles of muscle fibers. Bacterial cells were also found in the lumens of vessels.

In the 10–12 day interval, the histosections were observed to have pronounced fragmentary disintegration of muscle fibers. Almost no nuclei of cardiomyocytes and cells of the muscular connective tissue were found. The intermuscular connective tissue appeared as a non-structured mass, in which we saw colonies of bacteria (Fig. 8).

In the 13–15 day interval, we found no nuclei and alignment of cardiomyocytes in the muscle fibers. The intermuscular connective tissue was disintegrated, poorly stained. The collaged fibers were misaligned, fragmented.

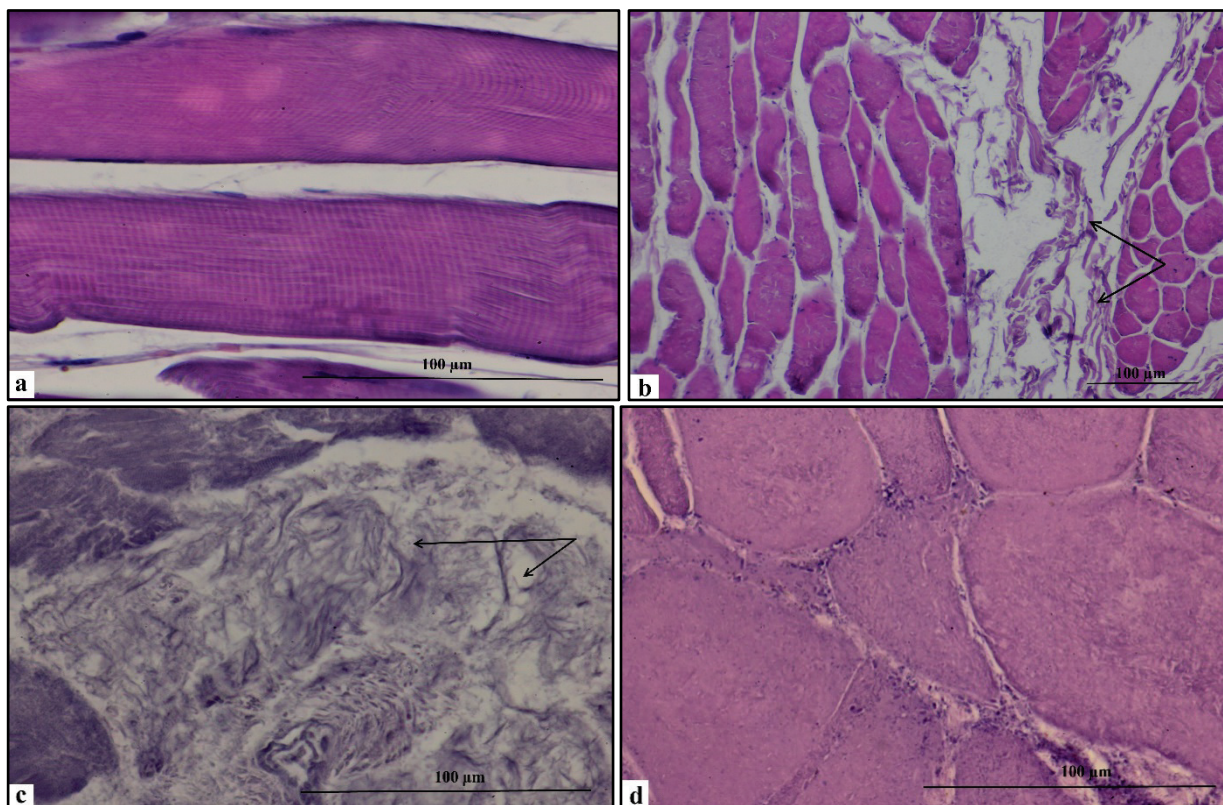
The microscopic structure of skeletal muscles in the 0–3 day postmortem interval corresponded to the histostructure of this organ in the norm (Fig. 9a). In the 4–6 day interval, nuclei in the muscle fibers were poorly stained. Alignment of the muscle fibers in some regions of the histopreparations was expressed poorly. The muscle fibers had a wavy pattern. In some areas, fragmentary breakdown of the muscle fibers was observed. In some regions, we saw disintegration and formation of transversal grooves. Collagen fibers were loose, misaligned, and of wavy configuration in some places (Fig. 9b). In some regions of the intermuscular connective tissue, we observed large bacterial colonies. In the 7–9 day interval, the muscle fibers completely lost their nuclei. No alignment of the muscle fibers was observed. The intermuscular connective tissue contained single bacterial cells (Fig. 9c) and colonies of bacteria.



**Fig. 8.** Cat myocardium, 10–12 day postmortem interval: detritus mass in the place of muscular connective tissue; hematoxylin and eosin

In the 10–12 day interval, changes in the muscle fibers were no different from such described on day seven. No nuclei in the muscle fibers were observed (Fig. 9d). Distinctive features were single completely ruined areas and large colonies of bacteria in the intermuscular connective tissue.

In the 13–15 day interval, we observed fragmentary and wad-like disintegration of the muscle fibers. In the intermuscular connective tissue, we observed some completely ruined regions. The connective tissue in some areas was a non-structured, wad-like substance. Large bacterial colonies were seen in the intermuscular connective tissue. In the 16–18 day interval, the muscle fibers were observed to have no nuclei, their wavy configuration was distinctly visible on the lengthwise sections. The muscle fibers have been undergoing fragmentary disintegration. We also observed large bacterial colonies.



**Fig. 9.** Skeletal muscles of a cat: *a* – 0–3 day postmortem interval, general view; *b* – 4–6 day postmortem interval, wavy configuration of the connective-tissue fibers; *c* – 7–9 day postmortem interval, bacterial cells in the intermuscular connective tissue; *d* – 10–12 day postmortem interval, absence of nuclei in the muscle fibers; hematoxylin and eosin

## Discussion

As of now, criteria of identifying time of death of agricultural and domestic animals, in particular domestic cat, have been developed insufficiently. Nonetheless, there is a variety of methods to identify time of death of humans (Sarkisova, 2021; Lytvynenko, 2022; Sokol, 2022; Voronov, 2022). Identifying postmortem interval according to histological changes, the method we proposed, has a number of advantages: the internal organs in animal corpses are less susceptible to the ambient factors and therefore are in better condition in found corpses for a certain time period since death, giving grounds for a more informative study (Delgado et al., 2021; Saber et al., 2021). Even if a corpse is missing some parts or organs, certain organs remain and can be used for the examination. For example, a common method of identifying time of death according to postmortem changes in the eyes (Garland et al., 2020; Perez-Martinez et al., 2020; Nioi et al., 2021) can be useless if the eyeballs are ruined (eyes are eaten by insects, pecked out by birds, etc.). The likelihood that a corpse has internal organs remaining is much higher. The collected and fixated samples, as well as histological preparations made of them, can be stored for a long time (Munmun et al., 2020). Such preparations can be used in the future as evidence in judicial cases, museum preparations, and visual material for education purposes, etc.

By using this method, visual materials can be made using common equipment in a quite short period of time, which helps in investigating cases related to death of animals. According to the results of our research, the method can be employed to identify the postmortem period for at least 18 days after death, thus having a good advantage over a number of other popular methods (thermometry, examination of livor mortis, etc). However, it also has a disadvantage – if an animal has been dead for more than 18 days, tissue disintegration reaches such a level that complicates the correlation of changes with time. Moreover, the condition of the tissues is negatively affected by the ambient conditions, and the internal organs could be pathologically altered. Nonetheless, its advantages are good reasons to use this method in forensic-veterinary and forensic investigations (Viciano et al., 2022), etc.

The microscopic structure of all the studied organs of domestic cat in the 0–3 day postmortem interval was consistent with such described in the literature sources (Qayyum, 1972; Yeager & Anderson, 1989; Williams et al., 1997; Mirhish et al., 2013; Cienfuegos et al., 2014; Leonard et al., 2021). The general patterns of changes in the tissues of the internal organs of cats are as follows: changes in the intensity of staining of cellular elements and intercellular structures on the histopreparations; formation of cavities filled with detritus; a number of distinctive changes in the connective-tissue stroma of the organs; emergence of rod-like bacteria and their colonies in the tissues.

Intensity of staining of certain cellular and tissue structures by the staining agents used in histological practice directly depends on the ability of certain responsive groups of chemical compounds that comprise the cells and tissues to form stable compounds with the stain molecules (Mohammed et al., 2023). Over time, macromolecule polymers disintegrate, which affects the ability of some structures to be stained. Therefore, with time, the ability of cellular and tissue elements to become stained by histological dyes decreases. Some structures can be completely unresponsive, poorly responsive, or unevenly responsive (vacuoles, cavities) to those stains.

Detritus is comprised of residues of cellular and tissue structures resulting from their disintegration. Histologically, detritus is homogenous, grainy, non-structured, poorly eosinophilous substance occupying a certain area in the field of view (Ceciliason et al., 2021). Formation and location of detritus can be of two types: distribution between still remaining structural elements of the tissue or emergence of clearly demarcated regions with detritus in places of certain structural tissue elements (Fig. 3). In the latter case, detritus can be either found on histopreparations or pass undetected due to peculiarities of the histopreparation making, and in such cases cavities are found instead of the regions with detritus. The observable amount of detritus and the area it occupies increases directly proportionately to time passed since death.

In the connective-tissue stroma of all the internal organs, besides loss of colour of nuclei, we observed emergence of the distinctive wavy pattern. In the norm, collagen fibers are usually straight. Wavy configuration

is obviously associated with ruination of fibrillar structure of collagen, brought about by autolysis and corpse decay. Collagen fibers progressively become loose, fragmented, and ruined, forming detritus. The waviest configuration of the connective-tissue fibers was in the 4–12 day interval.

Emergence of first bacterial cells and then bacterial colonies in the tissues of internal organs is related to contamination with microflora that has been present during the life, and which after death starts uncontrolled reproduction, facing no resistance from the defense systems of the body. We should note that emergence of bacteria and their colonies in the tissues of internal organs correlates with time past since death. At the same time, in the tissues that comprise the body surface, contamination with ambient microflora occurs, fostering the decay changes. This can lead to false interpretations and wrong conclusion on the time of death. Both single bacterial cells and their bacterial colonies of different sizes were found in the connective-tissue stroma of the liver, intermediate tissues of the liver and lungs, and the intermediate connective tissue. Bacterial cells were also often found in the lumens of the blood vessels. According to our research, this suggests the spread of putrefying microflora in the body through the blood circulatory system following death.

In the muscle tissue, both in the skeletal and in cardiac muscles, there were specific features according to which it is possible to identify time of death – loss of alignment of the muscular fibers and emergence of transversal grooves with following fragmentation. According to the authors, those changes are of autolytic nature.

Therefore, the development of the abovementioned traits in the tissues of the internal organs in corpses of animals correlates with time past since death, and therefore development of those features and its degree makes it possible to determine the postmortem interval (Delgado et al., 2021; Saber et al., 2021; Sokol, 2022).

## Conclusions

During the first 18 days after death, a number of postmortem histological changes developed in the examined organs of the domestic cats. They included changes in how intensively various cellular and tissue structures were stained by histological stains, disintegration of cellular and tissue structures into detritus, a number of changes in the connective tissues of the stroma of the organs, and emergence of bacteria and formation of their colonies in the tissues. Emergence and dynamics of development of the said changes, as revealed in our research, can be useful in identifying time passed since death within the indicated period and can be used in the practice of forensic veterinary medicine.

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