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MORPHOFUNCTIONAL CHANGES IN THE ORGAN OF IMMUNOGENESIS IN PARVOVIRUS AND CORONAVIRUS OF DOGS

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In rapid tests VetExpert and ELISA and PCR, changes in immunocompetent organs were identified, taking into account the peculiarities of the clinical manifestation of the disease and the results of macro- and microscopic changes in the organs and tissues of dogs with coronavirus and parvovirus enteritis.

Based on our analysis of literature sources, monitoring results and our own research, it was found that viral enteritis occupies a leading place in the infectious pathology of dogs and causes significant harm to animal owners. Viral enteritis leads to serious disorders of many body systems, and one of the first to be affected is the immune system.

Pathological dissection of dogs was performed by partial evisceration in the usual sequence. Prepared histological sections were stained with hematoxylin and eosin according to standard recipes. The general histological structure and microstructural changes of histo- and cytostructures of organs in histological samples were studied under a light microscope.

According to the results of pathological and anatomical autopsy of dogs in the intestinal form of parvovirus enteritis revealed changes in the immune system, namely: in the thymus most pronounced disorders of lymphocyte differentiation in the thymus, lymph nodes - dilation and overflow of blood vessels and edema of cortical and cerebral substances. swelling of the pulp and accumulation in the red pulp of granules and grains of iron-containing pigment - hemosiderin (due to the breakdown of a large number of erythrocytes).

With coronavirus enteritis in dogs, pathomorphological changes in immunocompetent organs are found, which characterize the suppression of immunogenesis during an infectious disease of viral etiology. Thus, the spleen has spotted hemorrhages, lymph nodes - moderately hyperplasia, with signs of hemorrhagic inflammation. Active proliferation of lymphoid cells, which leads to hyperplasia, is one of the markers of the pathogen's effect on the macroorganism in the form of an inflammatory process in regional lymph nodes, which indicates the multiplication of the virus and the development of immunological processes.

***Key words**: dogs, parvovirus enteritis, coronavirus enteritis, pathological and anatomical autopsy, macroscopic changes, histology, immune organs.*

Formulation of the problem. The problem of enteritis of viral etiology in animals is relevant for modern veterinary medicine. This is due to the widespread spread of such infections, especially among dogs, due to the increase in their population, which inevitably leads to an exacerbation of the epizootic situation, in particular, in relation to viral diseases. Therefore, in the general pathology of dogs enterovirus infections occupy a leading place [1, 2, 3].

In recent years, there has been an increase in the incidence of dogs with signs of diarrhea not only in Ukraine but also in Europe. During a set of laboratory studies, a significant prevalence of not only parvovirus but also coronavirus enteritis was found, which is extremely dangerous for puppies and dogs of small breeds due to rapid dehydration of their bodies and, consequently, death [4, 5, 6, 7].

Viral enteritis is the most common infectious disease of dogs, affecting the heart, liver, kidneys, intestines and more. The most common is parvovirus enteritis, but recently progressive and coronavirus, the causative agent of which is quite pathogenic for young animals and in the case of late diagnosis and treatment can be fatal [8, 9].

Analysis of recent research and publications. Today, the literature has accumulated the results of numerous immunological studies of materials, mainly blood, sick and healthy animals and humans. Most immunologists do not study the reactivity of a macroorganism on sectional material [10, 11, 12, 13].

No infectious disease in mammals can occur without protective and immunological reactions. Immunity, as the most important biological property of vertebrates, performs the function of protecting the genetic stability of the organism throughout life. Therefore, during the infectious process there is a

strong and intense immune response of the macroorganism to the pathogen. This determines the high sensitivity and specificity of immunological reactions [14, 15].

Given the relevance of this issue, the result of our study is to clarify, supplement and summarize data on the pathomorphology of various organs and tissues of dogs in coronavirus and parvovirus infection, which will determine the effect of the pathogen on animals in these pathologies.

Materials and methods of research. The aim of this study was to elucidate and characterize pathomorphological changes in immune organs, namely in the spleen, thymus and lymph nodes of dogs in parvovirus and coronavirus infections. The work was performed at the Faculty of Veterinary Medicine of Zhytomyr National Agroecological University (now Polissya National University), as well as in veterinary clinics of Zhytomyr, the study was based on the corpses of dogs of different breeds and sexes aged 2 to 8 months, who died with signs of infectious diarrhea. Only corpses of dogs with a clinical diagnosis of parvovirus enteritis (n = 15) and coronavirus enteritis (n = 9) were used for pathomorphological examination. Diagnostic tests to confirm coronavirus enteritis were performed using rapid tests VetExpert and in a private veterinary laboratory "Bald" (Kiev).

Pathological autopsy of dogs of different ages who died of coronavirus enteritis, was performed by partial evisceration in the usual sequence [16, 17]. For histological examination, the material (spleen, thymus, lymph nodes) after fixation in a 10% aqueous solution of neutral formalin was washed in running water, passed through alcohols of increasing strength and poured into paraffin. Histological sections were made of paraffin blocks on a sled microtome MS 2 with a thickness of not more than 10 μm. Dewaxed sections were stained with hematoxylin and eosin and applied to the balm according to generally accepted methods [18, 19].

The study is part of the research work of the Department of Anatomy and Histology, Faculty of Veterinary Medicine, Polissya National University: "Marker signs of immunogenesis and nervous system of vertebrates in onto- and phylogeny", № state registration 0120U102370.

Research results. At autopsy, coronavirus-killed puppies showed the same macroscopic changes in all animals, although they had minor individual differences. External examination of the corpses confirmed poor fatness, dullness, disheveled hair and dry skin, contamination of the tail root with fecal masses of yellow color.

The effect of the pathogen on the macroorganism usually leads to the development of secondary immunodeficiency, and the primary changes are structural changes in the thymus. In dogs with coronavirus infection, the thymus was flabby, swollen, and unevenly hyperemic. Histological examination of the thymus showed that all the blood vessels in the organ were dilated and filled with blood. The stroma of the connective tissue of the organ and the lobes of the thymus were clearly swollen.

No morphological changes in the capsule and trabeculae in the spleen of coronavirus puppies were recorded. However, there was swelling of the red pulp, the sinuses of this pulp were dilated, there was peritrabecular edema and lymph node hyperplasia. Lymphocytes in these nodules are usually solitary. The cells of their dense fibrous and smooth muscle tissue did not notice any noticeable changes. Isolated vessels with a damaged wall, from which homogeneous elements of blood flowed into the parenchyma of the organ. Macroscopic examination revealed that the organ itself was enlarged, sluggish, with isolated hemorrhagic infarcts of various sizes under its capsule, spotted hemorrhages and hyperplasia of the lymph nodes serving it.

Macroscopic changes were similar in all studied somatic and visceral lymph nodes. They had a whitish color, increased in volume (the capsule is tense, the connective tissue is fluffy, the lobular structure is pronounced, in the area of the parenchyma is pale pink, high humidity, the parenchyma protrudes above the capsule). Such macroscopic changes are characteristic of serous lymphadenitis. The lymph nodes of the small intestine are moderately enlarged, with signs of hemorrhagic inflammation.

The results of histological examinations revealed hyperplasia and hypertrophy of single clusters of lymphoid nodules of the submucosal basis of the mucous membrane of the small intestine. Lymphocytes in them were located mostly sparsely. In our opinion, such pathomorphological changes in the small intestine may be associated with metabolic disorders in coronavirus puppies in response to the pathogen, as evidenced by our histochemical studies (accumulation in the cytoplasm of enterocytes of basic and acidic proteins, hypersecretion of mucus cells). enterocytes chic-positive substances).

Hyperplasia (increase in the number) of lymph nodes was observed during morphological examination of intestinal lymph nodes in its cortical substance. Lymphocytes in them were located

sparsely, light centers are usually absent. However, in some of these nodules, mainly in their central part, there are foci of compacted lymphocytes. The cerebral substance is unevenly swollen, lymphocytes in it were located in separate groups, which included different numbers of cells. Among the lymphocytes were found monocytes and macrophages, located singly or in small groups. In some cases, arterial spasm was observed in the brain substance of various lymph nodes. As a result of our experiment, microscopic changes were detected in all somatic and visceral lymph nodes studied by us. No such changes were detected in the capsule. Capsule trabeculae, respectively, had no morphological changes.

In the pathological examination of the corpses of puppies that died from parvovirus, it was found that the corpses of the puppies were poorly fed. The skin and subcutaneous tissue were dry, indicating dehydration. Visible mucous membranes are bluish in color.

The spleen shrank slightly, causing the capsule to wrinkle slightly. The thymus is unevenly hyperemic and edematous, as a result of which the individual parts of the organ clearly increase and protrude beyond its common surface. All examined lymph nodes showed signs of serous-hemorrhagic lymphadenitis. They were enlarged (the capsule was tense, the parenchyma was swollen on the cut), high humidity, from the surface and on the cut had an uneven pink color.

In the thymus of dogs with parvovirus infection, all blood vessels of the parenchyma and stroma of the organ were clearly dilated and overflowing with blood. An uneven swelling of the connective tissue stroma of the organ was recorded between the lobules of the thymus. In addition, in most lobules of the thymus gland, a noticeable edema of their cortex and medulla was revealed, the cells of which were in a state of granular dystrophy. Some of the thymus bodies were disorganized, and some bodies were in a state of necrosis. Such changes in the histoarchitectonics of the thymus of dogs with parvovirus enteritis, in our opinion, may indicate a violation of the processes of lymphocyte differentiation in the organ.

When conducting histological studies of somatic and visceral lymph nodes, the nature of their microscopic changes was almost the same. However, the changes we found were more pronounced for the intestinal, hepatic lymph nodes and nodes of the pelvic cavity. The blood vessels of all somatic and visceral lymph nodes were dilated and filled with blood, their cortex and medulla oblongata were swollen. At the same time, its diffuse infiltration with a large number of erythrocytes was recorded in the medulla oblongata, but such infiltration did not appear in the cortex. Lymphoid nodules were poorly defined or absent. Single macrophages appeared in the lymph nodes, which in most cases were in close contact with lymphocytes. In the medulla of the lymph nodes, single macrophages were also found, which had already been in contact with both lymphocytes and altered erythrocytes.

During the histological examination of the spleen, its capsule was somewhat edematous, clearly manifested only at high magnifications of the microscope. The red pulp of the spleen was unevenly edematous, numerous foci of decay of a large number of erythrocytes were revealed in it, which led to the accumulation of granules and grains of iron-containing pigment - hemosiderin - in the intercellular space of the red pulp of the spleen, which was also manifested in large quantities in the cytoplasm of numerous macrophages (siderophages).

Lymphoid nodules in the spleen also experienced noticeable microscopic changes. Moreover, in some of the nodules, their borders were clearly delineated, due to which they were clearly differentiated in the red pulp. However, these nodules were located eccentrically relative to their central arteries. The other part of the lymphoid nodules was small in size with usually indistinct boundaries and poorly detected in the parenchyma of the organ. In most of these lymphoid nodules, a relatively large number of macrophages have appeared in the lymphocyte population, some of which are necrotic. Some lymphocytes, which were localized in lymphoid nodules and among other cells of the red pulp, contained eosinophilic inclusion bodies in their nuclei.

Conclusions. Pathomorphological changes of coronavirus infection in dogs in the available world literature are covered very superficially. We found that in coronavirus infection in dogs, microscopic changes in the organs of immunogenesis were registered: in the thymus – differentiation of lymphocytes in the cortical and cerebral lobes, uneven edema of the connective tissue stroma of the organ and severe cerebral edema and cerebral hyperplasia. lymphoid nodules. At the established complex of pathomorphological changes of intestines catarrhal-hemorrhagic inflammation was noted, and in the next lymph nodes – hemorrhagic inflammation. Such data, macro- and microscopic evaluation, indicate

inflammatory changes in the lymphoid organs – as a place of reproduction of the virus, which is an immunological process.

With parvovirus infection in dogs, microscopic changes in the organs of immunogenesis were recorded: in the thymus, a violation of the processes of differentiation of lymphocytes in the cortical and medulla layers of the lobules, uneven edema of the connective tissue stroma of the organ and pronounced edema of the cortex and medulla of the thymic lobules. The spleen is edematous, in the red pulp of the organ there was a significant amount of iron-containing pigment - hemosiderin, as a result of the breakdown of a large number of red blood cells.

Research prospects. To fully study the microscopic changes under the influence of parvovirus and coronavirus of dogs at the next stage it is advisable to investigate the features of this disease using histochemical methods.

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МОРФОФУНЦІОНАЛЬНІ ЗМІНИ В ОРГАНАХ ІМУНОГЕНЕЗУ СОБАК ЗА ПАРВОВІРУСНОГО ТА КОРОНАВІРУСНОГО ЕНТЕРИТУ

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У роботі, за допомогою експрес тестів VetExpert та в ІФА і ПЛР, визначено зміни у імунокомпетентних органах з урахуванням особливостей клінічного прояву хвороби та результатів макро- та мікроскопічних змін в органах і тканинах собак при коронавірусному та парвовірусному ентеритах.

На основі проведеного нами аналізу літературних джерел, результатів моніторингових та власних досліджень з'ясовано, що вірусні ентерити займають провідне місце в інфекційній патології собак і завдають значних збитків власникам тварин. Вірусні ентерити призводять до тяжких розладів багатьох систем організму, а однією з перших вражається – імунна.

Патологоанатомічний розтин собак виконували методом часткової евісцерації в загальноприйнятій послідовності. Виготовлені гістологічні зрізи фарбували гематоксиліном та еозином за стандартними прописами. Загальну гістологічну будову і мікроструктурні зміни гісто– та цитоструктур органів в гістологічних препаратах вивчали під світловим мікроскопом.

За результатами патолого-анатомічного розтину трупів собак за кишкової форми парвовірусного ентериту встановлено, зміни в органах імунного захисту, а саме: у тимусі найбільш виразними є порушення процесів диференціації лімфоцитів в тимусних часточках, в лімфатичних вузлах – розширення і переповнення кров'ю кровоносних судин й набряк коркової і мозкової речовини, в селезінці – набряк пульпи і накопичення в червоній пульпі гранул і зерен залізовмісного пігменту – гемосидерину, (внаслідок розпаду великої кількості еритроцитів).

За коронавірусного ентериту у собак виявляли патоморфологічні зміни в імунокомпетентних органах, що характеризують пригнічення функції імуногенезу під час інфекційного захворювання вірусної етіології. Так, у селезінці відмічаються крапчасті крововиливи, лімфатичні вузли – помірно гіперплазовані, з ознаками геморагічного запалення. Встановлено активну проліферацію лімфоїдного ряду клітин, що призводить до гіперплазії, і є одним з маркерів впливу інфекційного агента на макроорганізм у вигляді запального процесу в регіонарних лімфовузлах, що свідчить про репродукцію вірусу і вказує на розвиток імунологічних процесів.

Ключові слова: *собаки, парвовірусний ентерит, коронавірусний ентерит, патолого-анатомічний розтин, макроскопічні зміни, гістологія, імунні органи.*

МОРФОФУНЦИОНАЛЬНЫЕ ИЗМЕНЕНИЯ В ОРГАНАХ ИММУНОГЕНЕЗА У СОБАК ПРИ ПАРВОВИРУСНОМ И КОРОНАВИРУСНОМ ЭНТЕРИТЕ

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В работе, с помощью экспресс тестов VetExpert и в ИФА и ПЦР, определены изменения в иммунокомпетентных органах с учетом особенностей клинического проявления болезни и результатов макро- и микроскопических изменений в органах и тканях собак при коронавирусном и парвовирусном энтеритах.

На основе проведенного нами анализа литературных источников, результатов мониторинговых и собственных исследований установлено, что вирусные энтериты занимают ведущее место в инфекционной патологии собак и наносят значительный ущерб владельцам животных. Вирусные энтериты приводят к тяжелым расстройствам многих систем организма, а одной из первых поражается – иммунная.

Патологоанатомическое вскрытие собак выполняли методом частичной эвисцерации в общепринятой последовательности. Изготовленые гистологические срезы окрашивали гематоксилином и эозином по стандартным прописям. Общую гистологическое строение и микроструктурные изменения гисто- и цитоструктур органов в гистологических препаратах изучали под световым микроскопом.

По результатам патологоанатомического вскрытия трупов собак за кишечной формы парвовирусного энтерита установлено, изменения в органах иммунной защиты, а именно: в тимусе наиболее выразительными является нарушение процессов дифференциации лимфоцитов в тимусных дольках, в лимфатических узлах – расширение и переполнение кровью кровеносных сосудов и отек коркового и мозгового вещества, в селезенке – отек пульпы и накопления в красной пульпе гранул и зерен железосодержащего пигмента – гемосидерина, (вследствие распада большого количества эритроцитов).

При коронавирусном энтерите у собак проявляли патоморфологические изменения в иммунокомпетентных органах, характеризующие угнетение иммуногенеза при инфекционного заболевания вирусной этиологии. Так, в селезенке отмечаются точечные кровоизлияния, лимфатические узлы – умеренно гиперплазированные с признаками геморрагического воспаления. Установлено активное пролиферацию лимфоидного ряда клеток, что приводит к гиперплазии, и является одним из маркеров влияния инфекционного агента на макроорганизм в виде воспалительного процесса в регионарных лимфоузлах, что свидетельствует о репродукции вируса и указывает на развитие иммунологических процессов.

***Ключевые слова**: собаки, парвовирусный энтерит, Коронавирусная энтерит, патолого-анатомическое вскрытие, макроскопические изменения, гистология, иммунные органы.*