ORIGINAL ARTICLES | DISEASES OF SMALL PETS

ОРИГИНАЛЬНЫЕ СТАТЬИ | БОЛЕЗНИ МЕЛКИХ ДОМАШНИХ ЖИВОТНЫХ

DOI: 10.29326/2304-196X-2021-2-37-128-131 UDC 619:616.98:578.828.11:636.8

Prevalence of feline viral leukemia in Moscow and the Moscow Oblast with the analysis of hematological and biochemical blood parameters

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SUMMARY

Feline viral leukemia is an infectious disease that is common throughout the world. Despite the statistical records that have been successfully kept in other countries for many years, there is little data on the prevalence of the infection in the Russian Federation, and the samples are represented by a small number of animals. 11,807 blood samples collected in Moscow and the Moscow Oblast were tested for FeLV antigen and antibodies against FeLV by polymerase chain reaction, enzyme immunoassay, and immunochromatographic assay. The prevalence of feline viral leukemia was 12.8%, which correlates with the prevalence of the disease detected in developing countries, and speaks of a high disease prevalence in domestic cats in Moscow and the Moscow Oblast. Most often, feline leukemia virus was detected in free roaming domestic cats. During the diagnostic studies, the following hematological abnormalities were found: anemia, thrombocytopenia, lymphopenia, and a shift of the leukocyte formula to the left. Biochemical blood tests showed the increased levels of total protein, aspartate aminotransferase, alkaline phosphatase, and C-reactive protein. The obtained data demonstrate non-specific hematological and biochemical abnormalities in infected cats, and dictate the need for further study of the risk factors that predispose cats to the infection with this viral disease. Feline viral leukemia should be suspected in case of a non-specific clinical picture, as well as in case of abnormalities in blood biochemical and hematological parameters of free roaming cats or of those which have had a confirmed contact with their conspecifics.

Keywords: Feline viral leukemia, feline viral immunodeficiency, immunosuppression, prevalence, anemia.

Acknowledgements: The author would like to thank Vladimir V. Makarov, Doctor of Science (Biology), Professor of the Department of Veterinary Medicine of the ATI RUDN, for his help in writing and editing the article.

For citation: Zenchenkova A. P. Prevalence of feline viral leukemia in Moscow and the Moscow Oblast with the analysis of hematological and biochemical blood parameters. *Veterinary Science Today*. 2021; 2 (37): 128–131. DOI: 10.29326/2304-196X-2021-2-37-128-131.

Conflict of interest: The author declares no conflict of interest.

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УДК 619:616.98:578.828.11:636.8

Превалентность вирусной лейкемии кошек в условиях г. Москвы и Московской области с анализом гематологических и биохимических параметров крови

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РЕЗЮМЕ

Вирусная лейкемия кошек — инфекционное заболевание, распространенное по всему миру. Несмотря на статистический учет, который успешно ведется в других странах мира на протяжении уже многих лет, данные о распространении инфекции в Российской Федерации малочисленны, а выборки представлены небольшим количеством животных. Проведено исследование 11 807 образцов крови, собранных на территории г. Москвы и Московской области, на наличие антигена вируса лейкемии кошек и/или антител против возбудителя методами полимеразной цепной реакции, иммуноферментного и иммуно-хроматографического анализов. Превалентность вирусной лейкемии кошек составила 12,8%, что коррелирует с превалентностью заболевания, выявленной в развивающихся странах, и характеризует высокую степень распространенности инфекции в популяции домашних кошек г. Москвы и Московской области. Наиболее часто вирус лейкемии кошек выявляли у самцов и самок, имеющих свободный доступ на улицу. При проведении диагностических исследований установлены следующие гематологические отклонения: анемия, тромбоцитопения, лимфоцитопения и сдвиг лейкоцитарной формулы

влево. Биохимический анализ крови показал повышение уровней общего белка, аспартатаминотрансферазы, щелочной фосфатазы и С-реактивного белка. Полученные данные отражают неспецифичность гематологической и биохимической картины анализа крови, представленной у инфицированных кошек, и диктуют необходимость дальнейшего изучения факторов риска, предрасполагающих кошек к заражению данным вирусным заболеванием. Ветеринарные специалисты должны подозревать заболевание вирусной лейкемией кошек в случае наличия неспецифической клинической картины, а также в случае выявления отклонений от нормы параметров гематологического и биохимического анализа крови животных, имеющих свободный доступ на улицу или имеющих подтвержденный контакт с сородичами.

Ключевые слова: Вирусная лейкемия кошек, вирусный иммунодефицит кошек, иммуносупрессия, превалентность, анемия.

Благодарность: Автор выражает признательность доктору биологических наук, профессору департамента ветеринарной медицины АТИ РУДН Владимиру Владимировичу Макарову за помощь в написании и редактировании статьи.

Для цитирования: Зенченкова А. П. Превалентность вирусной лейкемии кошек в условиях г. Москвы и Московской области с анализом гематологических и биохимических параметров крови. Ветеринария сегодня. 2021; 2 (37): 128—131. DOI: 10.29326/2304-196X-2021-2-37-128-131.

Конфликт интересов: Автор заявляет об отсутствии конфликта интересов.

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INTRODUCTION

Feline viral leukemia is an infectious disease caused by feline leukemia virus (FeLV) belonging to the genus *Gammaretrovirus* of the family *Retroviridae*. This viral agent was discovered and described in 1964 by William Jarrett, and since then it has been found in blood and saliva samples from cats all over the world [1].

Feline leukemia virus is responsible for a wide range of clinical syndromes associated with immunosuppression and bone marrow disorders [2]. Thus, the clinical signs most often associated with immunosuppression are neutropenia, lymphocytopenia, and the development of such opportunistic infections as feline calicivirus infection, chlamydiosis, toxoplasmosis, and cryptococcosis [3]. Impaired bone marrow function leads to the development of non-regenerative anemia, which can be autoimmune, and thrombocytopenia. In addition, the risk of developing cancer (especially lymphoma) in infected cats is higher than in the healthy ones [4].

A large number of studies on the prevalence of feline viral leukemia in developed countries are presented in the literature sources. Thus, the prevalence of the infection in Southern Germany is 1.8% [5], in Switzerland – 3% [6], in the United States of America and Canada – 3.1% [7]. However, there is no exact data on the prevalence of feline viral leukemia in the Russian Federation due to the country's scale and the lack of necessary diagnostics. Nevertheless, in recent years, results of the studies aimed at determining the occurrence and prevalence of feline viral leukemia in certain cities of the Russian Federation have been published. Thus, the registered prevalence of feline viral leukemia in Vladivostok in 2018 was 15.9% [8].

Prevention plays a significant role in reducing the risk of virus transmission to healthy cats. The most effective measures include keeping the infected animals indoors, preventing them from contact with their conspecifics, mandatory castration of the infected animals, as well as vaccination against FeLV infection. Currently, non-adjuvanted recombinant and inactivated vaccines are available on the market, demonstrating the same effectiveness against the pathogen [9].

In connection with the above, the purpose of this research was to study FeLV prevalence in Moscow and the Moscow Oblast and to analyze abnormalities in blood biochemical and hematological parameters of the infected cats.

MATERIALS AND METHODS

Study design. 11,807 cat blood samples (both containing whole blood and serum) were tested for FeLV antigen and antibodies against FeLV. Data on animals and blood samples were collected between October 2018 and October 2019. 174 medical case histories were analyzed. The study included such criteria as gender, neuter status, access to the outdoors, and contact with conspecifics. The subject of the study was 6,529 male and 5,278 female cats.

Polymerase chain reaction. The study was based on the detection of the feline leukemia virus proviral DNA in blood of infected animals and was carried out using the Rotor-Gene Q amplifier (QIAGEN, Germany). Using a commercially available DNA extraction kit, QIAamp DNA Blood Kits (QIAGEN, Germany), proviral DNA was isolated from 200 µl of whole blood samples containing EDTA (Ethylene-diamine tetraacetate, anticoagulant). Amplification was performed using CFX96 Touch Real-Time PCR Detection System (Bio-Rad Laboratories, Inc., USA).

Enzyme-linked immunosorbent assay (ELISA). The study was aimed at detecting the feline leukemia virus antigen using SNAP FIV/FeLV Combo test system (IDEXX, USA) in whole blood samples containing EDTA and in the FeLV infected cat sera according to the kit instructions.

Immunochromatographic analysis (IHA). The study was based on the detection of feline leukemia virus antigen in blood serum samples using rapid test FIV Ab + FeLV Ag Combined Test (Quicking Biotech Co., Ltd., China) according to the manufacturer's recommendation.

Hematological and biochemical tests. The results of 124 general blood tests and 80 biochemical tests were assessed. Hematological testing of whole blood samples (25 μ l, containing EDTA) collected from infected cats was performed using an automatic analyzer Biocode-Hycel

Celly 70 (Biocode-Hygel, France). Biochemical testing of the infected cat blood sera (up to 40 μ l) was carried out using a BA-400 analyzer (BioSystems S. A., Spain).

RESULTS AND DISCUSSION

The study of 11,807 blood samples demonstrated 12.8% prevalence of FeLV (1,514 animals) in cats in Moscow and the Moscow Oblast. The obtained results correlate with the data on the disease prevalence in developing countries. For example, FeLV prevalence in Thailand is 16.5% [10], and in Brazil – 12.5% [11].

High FeLV prevalence in Moscow and the Moscow Oblast is probably due to the lack of recommendations for vaccination against this pathogen in the annual immunization schedule for healthy cats, as well as due to the large number of free roaming cats. According to the study, 89.7% of cats had their freedom to roam outdoors. Most often, feline leukemia virus was detected in male cats (61.3% – 928 animals), which may be due to their more aggressive zoo-social behavior towards their conspecifics, as well as their predisposition to live a feral-lifestyle. This observation confirms similar conclusions made in earlier studies [7]. In addition, 64.8% of cats (61 out of 94 animals) had a confirmed contact with their conspecifics at home or outdoors.

Despite the fact that some literature sources [2] provide data demonstrating that the risk of FeLV infection in non-castrated male cats is higher, the study did not prove it: 66.6% of the infected cats (618 animals) were neutered when the disease was diagnosed. Thus, the conclusion regarding the relationship between the neuter status of cats and the risk of infection with the feline leukemia virus in this study could not be made.

The summary of the results is presented in the Table.

Table
Prevalence of feline viral leukemia by sex, neuter status, and access to the outdoors

Распространенность вирусной лейкемии кошек в зависимости от пола, статуса интактности кошек и доступа на улицу

Parameter	Number of FeLV infected cats	Prevalence, %
Sex:		
male cats	928	61.3
female cats	586	38.7
Tota	l 1,514	12.8
Neuter status:		
neutered male cats	618	66.6
intact male cats	310	33.4
neutered female cats	411	70.2
intact female cats	175	29.8
Access to the outdoors:		
free-roaming	70	89.7
indoor keeping only	8	10.3
Tota	l 78	-
Contact with conspecifics:		
confirmed	61	64.8
absent	33	35.2
Tota	l 94	-

Assessment of 124 complete blood count test results revealed a decrease in red blood cell levels (RBC) in 46% of animals (57 cats), decreased hemoglobin levels (HGB) in 54.8% of animals (68 cats) and decreased hematocrit levels (HCT) in 60.5% of animals (75 cats). In addition, thrombocytopenia (PLT) was diagnosed in 58.1% of the tested cats (72 animals), an increased level of band neutrophils was found in 45.2% of cats (56 animals). Decreased lymphocytes levels were observed in 32.3% of cats (40 animals), and the increased erythrocyte sedimentation rate – in 37.9% of cats (47 animals).

Thus, the results obtained indicate that at the moment, the most common hematological abnormality occurring in infected cats in Moscow and the Moscow Oblast is anemia. Most often, it is of non-regenerative nature and can be caused by an autoimmune reaction. Therefore, diagnosis of FeLV infection should be included in the screening protocol for any cat with anemia signs.

It is worth noting that thrombocytopenia was diagnosed in more than half of the animals (58.1%), this fact can be associated with both aggregation of feline platelets induced by EDTA contained in test tubes for hematological testing, and an autoimmune reaction.

The following changes in the biochemical profile of the blood were observed during the studies: increased urea level – in 30% of cats, increased aspartate aminotransferase (AST) level – in 77.5% of cats, increased C-reactive protein level – in 87.5% of animals, hyperglycemia was observed in 56% of cats. In addition, water-electrolyte balance disorders were reported: hypokalemia and hyponatremia which may result from hypo- or anorexia were diagnosed 71.4% and 60% of cases, respectively, as well as some common disorders of the gastrointestinal tract, such as vomiting and diarrhea.

Thus, biochemical analysis of FeLV infected cat blood demonstrated non-specific results indicative of the pathologies not associated with the viral disease. It should be noted that most animals had abnormal blood biochemical parameters, typical for a non-specific inflammatory process: increased AST, C-reactive protein, and total protein levels.

CONCLUSION

Results presented in this study demonstrate a high prevalence of feline viral leukemia in Moscow and the Moscow Oblast. Based on the obtained from the medical case history of the infected animals, it can be concluded that the major risk factors associated with the disease are gender and access to the outdoors. Cats brought to a veterinary clinic had a wide range of clinical manifestations associated with viral leukemia. The most pronounced abnormality in the hematological parameters in infected cats, which may be a good reason to suspect feline viral leukemia, was anemia. We believe that further research is needed on the prevalence of feline viral leukemia in the Russian Federation, the risk factors, and the clinical signs associated with this viral disease.

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Received on 29.01.2021 Approved for publication on 04.03.2021

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