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Biocoenotic Diagnostics of Unfavorable Factors in the Cows Infection of Farms in the Moscow Region

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ABSTRACT

The article presents data of detailed biocoenotic diagnostics in 12 farms of the Moscow region affected by obstetric-gynecologic diseases of cows (endometritises, and mastitises), as well as gastrointestinal and respiratory diseases of newborn calves with the aim of identifying zoo-hygienic, zootechnical and veterinary deficiencies to optimize a combat strategy against factor infections. To this end, periodic visits were made to farms for epizootological examinations of farm biogeocenoses and the selection of blood serums, heparin-stabilized blood and samples of pathological material for bacteriological, mycological, virological, hematological and immunological studies.

An analysis of the epidemiological situation for cattle diseases caused by conditionally pathogenic bacteria shows that in farms of the Moscow region, where factor infections are unfavorable, there are conditions for the circulation of pathogens, multiple passage through susceptible animals, increased virulence of conditionally pathogenic microorganisms, and in some cases, the emergence of pathogenicity in saprophytic bacteria. It is shown that the main factors contributing to the development and dissemination of these pathologies are the unsatisfactory food basis of farms, violations of elementary veterinary and sanitary rules for keeping and milking, failure to fully implement measures to prevent these diseases due to the lack of sufficient material resources in the farms. All the above demonstrates that experienced farms are lacking adequate conditions to ensure the epidemiological chain in the infectious process.

Effective control of malignant microbial ecosystems require in-depth knowledge of the quantitative and species composition of parasitocenoses, the study of relationships between its individual representatives, required isolation of pure cultures of all his affiliates, the determination of marker biological properties of microbial agents, and the conduct of a detailed biocenotic diagnosis of the animal habitat (farming, barn, etc.).

Keywords: Farming, Biogeocenosis, Cows, Keeping, Feeding, Milking Regime, Factor Infections.

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INTRODUCTION

Ensuring reliable food security of the country is currently one of the most important state tasks. Therefore, the tasks set by veterinary medicine, namely: optimizing veterinary services, reducing the incidence and mortality of animals, play a significant role in improving the quality of life of the whole society.[1,2,3] However, despite the disaggregation of cattle farms during the reform of agriculture, factor infections caused by associations of conditionally pathogenic microorganisms in farm animals remain the number one problem for veterinary medicine specialists. This is due to the fact that in reorganized livestock farms the traditional technology of milk production is applied, biogeocenoses preserved in the process of evolution, including associations of conditionally pathogenic bacteria that cause various pathological processes in animals under farm conditions, have been preserved.[4,5,6,7,8,9,10,11,12,13]

The underestimation of microorganisms as living beings capable of complex relationships has led to the fact that there are currently no clinical recommendations that allow clear regulation of the actions of a veterinarian aimed at fighting diseases caused by associations of microorganisms. If the veterinarian does not take into account at least one of the parasitocenosis joints that is involved in the etiology of the disease, this leads to a significant decrease in the effectiveness of control measures that are carried out on the farm.[44,45,46,47,48,49,50]

Livestock farms should be considered as biogeocenoses artificially created by human. The underestimation of microorganisms as living beings capable of complex relationships has led to the fact that there are currently no clinical recommendations that allow clearly regulating the actions of a veterinarian aimed at fighting diseases caused by associations of microorganisms. If a veterinary specialist does not take into account at least one of the joints of parasitocenosis, which takes part in the etiology of the disease, this leads to a significant decrease in the effectiveness of control measures that are carried out in the farm.[14,15,16,17,18,19,20]

Livestock farms should be considered as man-made biogeocenoses. In them there are special relationships between animals, helminths, protozoa, and various microflora, which are radically different from those in natural conditions. In artificial biogeocenoses, conditionally pathogenic microflora that circulates in the farm can cause various associated diseases in farm animals. These are diseases such as mastitis, endometritis, vaginitis in adult

animals and gastrointestinal and respiratory diseases in newborn young animals.[5,21,22,23,24,25,26,27,28,29,30,31,32]

An important step of the diagnostic process is the assessment of the farm biocenosis in which the disease occurred. In this case, it is necessary to take into account the type of dysfunctional biocenosis, its features, the presence of geochemical zones according to the results of studies of soil, feed and a number of endemic diseases, which in turn can lead to a decrease in the resistance of the animal organism. When evaluating farmers's biocenoses, they must pay attention to the conditions of keeping, microclimate and food chains. Adverse changes in the microclimate can cause metabolic disorders and diseases caused by conditionally pathogenic microflora. When analyzing food chains, attention is paid to all links in the production, storage and preservation of feed, since these processes affect their chemical composition and, ultimately, the health of animals. Importance should also be given to assessing the bodies of water that are used for watering animals. A biocenotic diagnosis makes it possible to predict the disease, which is important for the development of preventive measures. Anti-epizootic measures that are carried out with the data of the assessment of the biocenosis in which animals of the disadvantaged herd live have a great economic efficiency.[5,6,10,33,34,35,36]

For definitive diagnosis usually requires laboratory studies including virological, bacteriological, mycological and parasitological studies. Serological methods of diagnosis are important, if not decisive, and in some cases, histopathological studies are required to identify pathognomonic signs. An integrated approach to the diagnosis of parasitocenosis has the advantage that if the results obtained by one method are unclear, this gap can be filled by other research methods.[20,34,37,38,39,40,41]

The elements of biocenotic diagnostics, which must be applied in the fight and prevention of animal diseases, are the key to increasing their productivity, environmentally sound production of high-quality products for human society as part of its sustainable development. Therefore, the study of the epidemiological situation of diseases caused by conditionally pathogenic bacteria, it is important biocenotic diagnostics, i.e. the analysis of conditions, maintenance and feeding of animals, as well as an analysis of the followers of microbial associations animal farm, circulating in some farms and cause various diseases in animals. Based on the above, conducting a detailed epidemiological assessment of biocenoses of farms in the Moscow region that are unfavorable for factor infections is an important direction for scientific research.

OBJECTIVE

To conduct a detailed biocenotic diagnostics of 12 farms in the Moscow region that have problems with obstetric and gynecological diseases of cows, as well as gastrointestinal and respiratory diseases of newborn calves in order to identify zoohygienic, zootechnical and veterinary deficiencies in order to optimize the strategy for fighting factor infections.

MATERIALS AND METHODS

Study of biocenoses were conducted in 12 farms in the Moscow region, which were not affected by the development of factor infections in farm animals (obstetric and gynecological diseases of cows, gastrointestinal and respiratory diseases of newborn calves) with a total number of 12254 cattle, including 4445 cows. For this purpose, field trips were periodically carried out for epizootological examinations of farm biogeocenoses and the selection of blood serum stabilized with heparin and samples of pathological material for bacteriological, mycological, virological, hematological and immunological studies.

Criteria for inclusion in the study: disadvantaged farms for respiratory and gastrointestinal diseases in newborn calves, as well as for obstetric and gynaecological diseases of cows.

Criteria for inclusion in the study: dysfunctional farms for respiratory and gastrointestinal diseases in newborn calves, as well as obstetric and gynecological diseases in cows.

Exclusion criteria: farms that are safe from factor infections; incomplete clinical, laboratory and pathoanatomic data.

In order to ensure the confidentiality of information that may contain trade secrets, the farms in which the studies were conducted were distributed and encrypted using envelopes MTF1-MTF12 (MTF - dairy farm).

During the epidemiological survey of farms, first of all, the incidence of cows was studied with obstetric and gynecological diseases, gastrointestinal and respiratory diseases of newborn calves. Attention was also drawn to the presence or absence of other infectious diseases in the farms (tuberculosis, leukemia, acute respiratory diseases).

The exclusion of the most widespread viral diseases in cattle (infectious rhinotracheitis and parainfluenza) in experimental farms was carried out by serological methods twice with an interval of 14 days on the basis of the laboratory of clinical research methods in the department of veterinary medicine of the RUDN Agricultural Research Institute. To detect antibodies to the parainfluenza-3 virus in cattle, a hemagglutination inhibition reaction was used. To set up the reaction, we used the "Set for diagnostics of bovine parainfluenza-3" in the reaction of inhibition of hemagglutination of RTGA (TU-10-19-84-89) produced by LLC "AgroVet" LLT (Moscow). The reaction was carried out according to the protocol recommended by the manufacturer. The titer of specific antibodies to the causative agent of infectious rhinotracheitis of cattle was determined in the indirect hemagglutination reaction according to the standard method using the Agrovet LLC kits: a erythrocyte diagnostic kit for serodiagnosis of infectious rhinotracheitis in cattle in the indirect hemagglutination reaction. Microplanes were used for serological reactions.

Carrying out a diagnostic examination of the biocenoses of livestock farms, the conditions of keeping, feeding and operating animals were analyzed in detail. The temperature and humidity regime, speed of movement and gas composition of the air in the premises of livestock farms were studied using generally accepted zoohygienic methods. On dairy farms, temperature and air humidity were determined with a TKA-PKM combined device (model 42),

air movement speed was determined with a TKA-PKM hot-wire anemometer (model 50), the carbon dioxide content in the air was measured according to Hess, ammonia and hydrogen sulfide were universal gas analyzer UG-2, microbial contamination and dust content in the room air - by the Krotov apparatus. The parameters of the microclimate in livestock premises were taken into account three days in a row in three zones: in the middle of the room, in the corners of the ends diagonally with the calculation of arithmetic averages.

In order to evaluate the adaptation characteristics of animals, the main clinical and physiological indicators were studied: pulse frequency, respiratory movement frequency and body temperature according to conventional methods in veterinary science. Animal's behavioural characteristics were studied by the method of time-based observations.

In order to evaluate the adaptation characteristics of animals, the main clinical and physiological indicators were studied: pulse frequency, respiratory movement frequency and body temperature according to conventional methods in veterinary science.

Bacteriological, mycological and hematological studies, and also define some properties of isolated cultures of bacteria were performed in the laboratory of clinical research methods in the veterinary medicine Department of the Agrarian Technological Institute of RUDN.

Milk samples were taken from cows with mastitis, cervical exudate from endometritis, and pieces of internal organs and lymph nodes from fallen newborn calves. Milk samples for studies on mastitis were passed into sterile test tubes; samples of exudate from the cervix for endometritis were taken with sterile cotton swabs into sterile test tubes. Before sampling the milk, the udder teats were wiped with a swab dipped in 70° ethanol. The first portion of milk (5-10 cm3), which was located in the nipple channel, was put in a separate dish. For microbiological analysis were taken subsequent portions of milk. Before selecting mucus from the cervix, the researcher's hands and labia were treated with 70° ethanol. An assistant over the open flame of the alcohol lamp opened a sterile bacteriological tube with a tampon and handed it to the researcher, who with one hand expanded the labia, and with the other – carefully inserted a sterile tampon into the vagina. Newborn calves that were killed or forcibly slaughtered for diagnostic purposes were subjected to a pathoanatomic autopsy and selected pieces of internal organs (spleen, liver with gall bladder, lungs, kidneys, as well as mesenteric and middle mediastinal lymph nodes) according to generally accepted methods of selecting pathological material. The pathologic material was placed in sterile plastic bags and was subjected to microbiological studies no later than 6 hours after sampling directly at the farm or in the laboratory.

During microbiological studies, the selected pathological material was used for culture media with a Pasteur pipette. For yeast-like fungi, Saburo medium was used, staphylococci-peptone-salt medium, yolk-salt agar, meat-peptone agar, enterobacteria-Endo medium, Ploskirev medium, bismuth-sulfite agar. The crops were again incubated in a thermostat at 37-38° C for 24 hours, and in the absence of growth, the cups were kept for up to 3 days.

After studying the cultural and morphological properties of all individual type colonies, they were transplanted into test tubes and incubated at 37-38 °C for 24 hours. The resulting pure bacterial cultures were tested for mobility in crushed droplet preparations using phase-contrast microscopy in a darkened field of view and subjected to identification.

The morphology of bacteria was studied in smears stained by Gram and Romanovsky-Giemsa. Further identification by biochemical properties was carried out in accordance with the "Bergey bacteria Determinant".

Gram-negative rods that gave a positive result in the test for the presence of catalase, negative-in the test for cytochrome oxidase, oxidized and fermented glucose (in the Hugh-Leifson medium), reduced nitrates were referred to the Enterobacteriaceae family.

Was performed by inoculation of all the isolated cultures in the environment Giss with glucose, maltose, lactose, mannose, sucrose, mannitol and dulcitol. Gram-positive rod-shaped bacteria were additionally transplanted to the Gissa medium with galactose, salicin, fructose, and arabinose. In order to determine the catalase activity of microorganisms, the bacterial mass removed from the agar surface by the loop was suspended in a drop of 3% hydrogen peroxide on the slide.

To further identify members of the Enterobacteriaceae family to genus and species, the culture was seeded on Olkenicki medium, a long pesky row that included media with mannite, maltose, sucrose, xylose, ramnose, dulcite, sorbitol, salicin, fernet salt (d-tartrate), milk with lacmus, and acetate recovery, also carried out on the indole test, H_2S the presence of methyl.

In gram-negative rod-shaped bacteria (bacillus), the fermentation of carbohydrates such as inositol and sorbitol, utilization of citrate and sodium malonate, production of hydrogen sulphide, indole and acetylmethylcarbinol, and the presence of ornithine decarboxylase, lysine decarboxylase, phenylalanine deaminase and β-galactosidase were additionally determined using indicator paper systems (IRB) (N. Novgorod). To eliminate mobility in cultures of the genus Proteus, 96° alcohol was poured into bacteriological cups with MPA before conducting research, kept for 3-5 minutes, and then the alcohol was removed.

Determination of serovariants isolated from pathological material of Salmonella cultures was carried out in the agglutination reaction on glass using a set of Salmonella O-complex and monoreceptor O - and N-agglutinating sera of Biofactory production in accordance with the instructions. Determination of E. coli serogroups was performed using a set of "serum "O"-coliaglutinating" (FSUE "Armavir Biofactory"). To identify the bacteria of the Pseudomonadaceae family (Pseudomonas), the culture was transplanted to the king medium, in a test tube with BCH and grown in a thermostat at a temperature of 42° C. The presence of catalase was determined to differentiate Staphylococcus bacteria from the Streptococcus genus. To differentiate the genus Staphylococcus from the genus Micrococcus, a glucose oxidation-fermentation test (Hugh-Leifson medium) was used. To identify bacteria of the genus Staphylococcus, species were tested for the presence of coagulase, oxidation of mannitol, galactose, maltose, lactose,

sucrose; growth ability in the presence of 10% NaCl. To identify bacteria of the genus Streptococcus before species, tests were performed on the ability to grow in air, at 10° C and 45° C, pH 9.6, in the presence of 6.5 % NaCl, 40 % bile; hemolysis; fermentation of sugars.

To determine the pathogenicity of isolated cultures, three white mice weighing 14-16 g were administered intraperitoneal 1 billion microbial cells for each strain of the microorganism. Laboratory animals were observed for 5 days. Culture was considered pathogenic in case of death of one or more of the mice for five days after infection. At the death of the animal, conducted bacteriological examination of the blood (the coloration by the Gram, Romanovsky-Giemsa, and Burri-Gins).

The obtained research results were processed statistically and presented in the form of tables.

RESULTS AND DISCUSSION

As indicated in the section "Materials and methods", research on the study of microbiocenoses was carried out in 12 farms of the Moscow region, which were selected on the basis of inclusion-exclusion criteria. All 12 farms in which we conducted research are located evenly across the entire territory of the Moscow region.

A detailed description of the biocenoses of livestock farms, among the animals that we have studied, is given below in TABLES 1-6.

Table 1: Number of cattle in experimental livestock farms

Farms in Moscow's region	Animal's population	
	Total	Cow
Farm MTF1	1324	452
Farm MTF2	2208	696
Farm MTF3	524	147
Farm MTF4	486	257
Farm MTF5	1527	748
Farm MTF6	1412	571
Farm MTF7	689	156
Farm MTF8	1089	360
Farm MTF9	643	192
Farm MTF10	697	264
Farm MTF11	425	146
Farm MTF12	1230	456
Total	12254	4445

In total, 12254 heads of cattle, including 4445 cows, were kept in farms where we studied microbial associations of opportunistic bacteria and the relationship between individual joints of bacterial parasitocenoses. From the data shown in (TABLE 1), it is also clear that the number of cows in the experimental disadvantaged farms ranged from 146 to 748 heads. It should be noted that farms in the Moscow region mainly contain animals of the red steppe, black-and-white and Simmental breeds. In experimental farms where epizootological studies were conducted (except for MTF 2, MTF 12, where Simmental breed animals are bred), red steppe cattle are kept.

Every living being needs to receive nutrients into the body. Conditions for keeping and feeding cows in experimental farms are shown in (TABLE 2).

Table 2: Conditions for keeping and feeding cows on farms

Farm	Stream	Presence		Veterinary and sanitary condition of premises	Feed supply of the farm, %	Feeding diet for cows
		Maternity wards	dispensaries for calves			
MTF_1	Breed	+	+	Satisfying	94	Silage- 14kg; hay-4kg; straw-3kg; roots-2kg; compound feed-2kg
MTF_2	Breed	+	+	Satisfying	88	Silage-12kg; hay-1,4kg; straw-1,7kg; compound feed-4kg
MTF_3	Tradable	––	––	Not satisfying	86	Silage-15kg; hay-2kg; straw-5kg; compound

						feed-2kg; haylage-5kg
MTF_4	Tradable	––	––	Not satisfying	67	Silage-11kg; straw-3kg; corn- fed-2kg; oilcake-1,5kg
MTF_5	Tradable	1	––	Satisfying	70	Silage-15kg; hay-3kg; straw-6kg; grist-1kg
MTF_6	Tradable	––	––	Not satisfying	73	Silage-10kg; hay-3kg; straw-2,5kg; compound feed-4,5kg
MTF_7	Tradable	1	––	Not satisfying	79	Silage-9kg; hay-2,5kg; straw-4kg; canned food-1,5kg
MTF_8	Tradable	––	––	Satisfying	98	Silage-22kg; hay-3kg; straw-1kg; corn- fed-1kg; macukha-0,5 kg
MTF_9	Tradable	––	––	Not satisfying	78	Silage-14kg; hay-6kg; straw-3kg; macukha-100g for 1l milk
MTF_{10}	Breed	+	––	Satisfying	86	Silage-15kg; hay-1kg; straw-5kg; macukha-0,7 kg
MTF_{11}	Tradable	––	––	Not satisfying	72	Silage-13kg; straw-4kg; grist-1,5kg; compound feed-2kg
MTF_{12}	Tradable	1	––	Not satisfying	74	Silage-12kg; hay-1,8kg; straw-2kg; compound feed-2kg; macukha-0,5 kg

Note: + — are present; - — lack; 1 — one non-replaceable maternity ward

An analysis of animal conditions and feeding shows that all farms do not fully provide livestock with feed. In only five farms (MTF1, MTF2, MTF3, MTF8, MTF10) the supply of fodder exceeded 80%. Therefore, to fully meet the needs of livestock farming experienced farms, their owners were recommended the following measures: to expand the area of cultivation of perennial (alfalfa, esparcet, donut, Wheatgrass) and annual grasses (spring rape, oilseed radish), as well as multi-crop crops (sudang, sorghum), which will significantly reduce the cost of feed by 40-50% and increase their biological value; improve natural forage lands by land reclamation, sowing of various grasses, drainage swampy areas; to constantly water unused areas of arable land; to restore and improve winter pastures by sowing oats, oat-pea mixture, spring rape and other crops in summer; to significantly increase the production of silage crops, herbs for hay, and root crops on arable land.[42,43,44,45,46,47]

It should be noted (TABLE 2) that three farms (MTF1, MTF2, MTF10) have a breeding direction of cattle breeding, the remaining 9 farms are engaged only in the production of milk, that is, they have a commodity direction. It should also be noted that only three farms (MTF1, MTF2, MTF10) had replacement maternity units, and three more farms (MTF5, MTF7, MTF12) had one non – replaceable maternity unit. Almost all farms (except MTF1, MTF2) did not have replacement dispensaries for keeping newborn calves. All of the above indicates that experienced farms do not have the appropriate conditions to ensure that the epidemiological chain is broken during the infectious process.

Indoor microclimate during the long winter-stall period, the periodic change of day and night are the main factors that determine the rhythm and intensity of physiological processes in the body, as well as have a significant impact on the growth and development of animals. Research on the study of zoohygenic parameters of premises where animals were kept in experimental farms was conducted in January – the most severe period of wintering (measurements were made 1 time a day for a month). Microclimatic indicators of MTF1-MTF12 premises are presented in (TABLE 3).

Table 3: Environmental indicators in the areas of farms

Farm	Microclimate indicators				
	Temperature °C	Relative humidity,%	Contents co2,%	Ammonia content, mg/l	Air speed,m/sec
MTF_1	5.6±0.12	87.67±1.24	0.10±0.02	0.011±0.01	0.26±0.06
MTF_2	3.2±0.22	82.34±0.98	0,12±0,01	0.014±0.01	0.23±0.09
MTF_3	4.7±0.11	85.53±1.31	0,14±0.02	0.013±0.01	0.28±0.04
MTF_4	2.1±0.08	64.13±2.45	0.21±0.03	0.024±0.02	0.42±0.05

MTF_5	2.4±0.13	68.32±2.11	0,17±0.03	0.027±0.02	0.34±0.04
MTF_6	3.2±0.09	74.41±1.54	0,20±0.02	0.016±0.01	0.31±0.06
MTF_7	1.3±0.04	61.33±3.65	0,24±0.04	0.025±0.02	0.55±0.15
MTF_8	2.8±0.07	73.11±1.63	0,18±0.03	0.015±0.01	0.40±0.12
MTF_9	3.7±0.18	84.24±1.14	0,17±0.02	0.014±0.01	0.26±0.08
MTF_{10}	1.6±0,09	67.56±5.78	0,22±0.03	0.018±0.01	0.42±0.11
MTF_{11}	1.1±0.06	62.32±4.32	0,18±0.02	0.023±0.02	0.47±0.19
MTF_{12}	2.3±0,12	70.31±1.43	0,15±0.01	0.020±0.02	0.35±0.10

The established variations in the microclimate in the surveyed areas are explained by drafts, imperfections in air exchange, and lack of organized regulation of supply and exhaust ventilation.

The veterinary and sanitary condition of premises in almost all farms was unsatisfactory. This is due to the fact that manure removal from premises is carried out in most farms once or twice a day. For several years, some farms do not carry out preventive and technological disinfection. All this contributes to the accumulation of conditionally pathogenic bacteria in the premises and creates conditions for faster passage of microorganisms and increasing their virulence.

Analyzing the behavior of animals in the premises of farms (MTF1, MTF3, MTF6 and MTF9), it should be noted that the temperature in buildings and relative humidity of the air were beneficial to animals, they spent more time in motion and ate food, since active movement increases metabolic processes in the body, which contributes to increased appetite, greater feed consumption.

It should also be noted that almost all farms observed violations in the ventilation of premises, in feeding dry cows, as well as drinking the first portions of colostrum to calves, which was carried out with a long delay (up to 5-8 hours). In addition, it was found that all experienced farms do not have diagnostic tests of cows in the first days after calving for subclinical mastitis. This fact, in our opinion, significantly affects the reduction of animal resistance and creates favorable conditions for colonization of the gastrointestinal tract of newborn calves with conditionally pathogenic microflora, which causes the development of subclinical mastitis in cows.

The analysis of compliance with milking regimes in experienced farms found its imprint in (TABLE 4).

Table 4: Cow milking practices in farms

Farm	Method of milking cows	Milking rate	Observation of mechanical milking mode	Compliance with veterinary and sanitary rules when milking cows
MTF1	Mechanical	3 times	Vacuum failure	Respected
MTF2	Mechanical	2 times	Vacuum failure	Respected
MTF3	Manual	2 times	––	Respected
MTF4	Manual	3 times	––	some violations
MTF5	Mechanical	3 times	Respected	Respected
MTF6	Mechanical	3 times	Not respected	some violations
MTF7	Manual	2 times	––	There are serious violations with individual milkmaids
MTF8	Mechanical	2 times	Vacuum failure	Respected
MTF9	Manual	2 times	––	There are serious violations with individual milkmaids
MTF10	Manual	2 times	––	There are serious violations with individual milkmaids
MTF11	Manual	2 times	––	Respected
MTF12	Mechanical	3 times	Not respected	Some violations

Note : - — mechanical milking is not holding

Milking cows in all experimental farms (TABLE 4) is carried out by machine and manual methods 2-3 times a day. In cases, we have identified significant shortcomings in compliance with the veterinary and sanitary rules of milking. These violations create conditions for increasing the incidence of cows with mastitis and, as a result, lead to malignant colonization of the intestinal tract of newborn calves with microflora, which leads to the development of dysbiosis and gastrointestinal disorders caused by conditionally pathogenic bacteria.

The physiological functions of the animal body depend directly of the environmental conditions. Since the existence of animals is inseparable from the environment, and changes in environmental conditions can affect changes in the human body. The study of such physiological indicators as body temperature, pulse rate and respiration allows us to judge the state of health of the animal and its adaptive capabilities.

Data from clinical studies showed that experimental animals from all experimental farms did not have significant deviations in the physiological functions of the body.

A detailed analysis of the epidemiological situation for infectious diseases of cattle in experimental farms is presented in (TABLE 5).

Table 5: Epidemiological Situation cattle's Infectious Diseases

	Presence of infectious diseases caused by obligate pathogens	Conditionally pathogens were isolated	
		by veterinary medicine labs in previous years	By us during 2018-2020
MTF1	Absent	*E. coli* (O8, O111)	*E. coli* (O8, O9, O111), *P. aeruginosa*, *P. vulgaris*, *S. epidermidis*, *S. agalactiae*, *S. faecalis*, *S. saprophyticus*
MTF2	Absent	*E. coli* (O1, O126), *P. aeruginosa*	*E. coli* (O1, O126), *P. aeruginosa*, *K. pneumoniae*, *S. aureus*, *S. dysgalactiae*
MTF3	Absent	*E. coli* (O8), *S. dublin*	*E. coli* (O8, O22), *S. dublin*, *P. vulgaris*, *S. agalactiae*, *S. faecalis*, *S. intermedius*
MTF4	Absent	*E. coli* (O9), *S. enteritidis*	*E. coli* (O4, O9, O126), *P. aeruginosa*, *P. vulgaris*, *S. aureus, S. uberis*
MTF5	Absent	*E. coli* (O8), *S. enteritidis*	*E. coli* (O4, O8, O101), *P. vulgaris*, *C. albicans*, *S. aureus*, *S. uberis*, *S. saprophyticus*
MTF6	Absent	*E. coli* (O9)	*E. coli* (O4, O9, O18), *P. aeruginosa*, *P. vulgaris*, *S. aureus*, *S. uberis*, *S. pyogenes*
MTF7	Absent	*E. coli* (O8)	*E. coli* (O8, O78), *P. aeruginosa*, *P. vulgaris*, *B. subtilis*, *S. aureus*
MTF8	Absent	*E. coli* (O111)	*E. coli* (O26, O33, O111), *S. aureus*, *S. agalactiae*, *S. uberis*, *C. albicans*
MTF9	Absent	*E. coli* (O8), *S. enteritidis*	*E. coli* (O8), *P. aeruginosa*, *S. enteritidis*, *P. mirabilis*, *S. aureus*, *S. pneumoniae*, *S. pyogenes*, *S. saprophyticus*
MTF10	Absent	*E. coli* (O4, O101), *P. aeruginosa*	*E. coli* (O2, O4, O101), *P. aeruginosa*, *P. mirabilis*, *P. multocida*, *S. aureus*, *S. dysgalactiae*, *S. pneumoniae*
MTF11	Absent	*E. coli* (O8, O33), *S. dublin, S. uberis*	*E. coli* (O8, O33), *P. aeruginosa, S. saprophyticus, S. pneumoniae, S. aureus, S. uberis*, *P. mirabilis*
MTF12	Absent	*E. coli* (O9, O26), *S. enteritidis*	*E. coli* (O9, O26, O78), *K. oxytoca*, *S. typhimurium*, *S. pyogenes*, *S. uberis*

An analysis of the epidemiological situation found that all farms where we conducted research, infectious diseases caused by obligate parasites were not registered. An analysis of the results of bacteriological studies carried out in previous years by specialists of the laboratories of veterinary medicine of the Moscow region and us during 2018-2020 shows that in previous years (until 2018), similar microflora was practically isolated by us. At the same time, it is clear that there is also some evolution of the microbial landscape that circulates in farms.

As shown in (TABLE 5), the variability is affected by changes in both the number of microorganisms in microbial associations, and their serovars and serotypes. In our opinion, the broader microbial landscape we have found is also related to the fact that some veterinary medicine laboratory workers believe that gastrointestinal and respiratory diseases of newborn young are caused by a single pathogen, and not by an Association of opportunistic bacteria. At the same time, bacteriological studies can only be reduced to isolation of a single pathogen, which does not give a complete picture of the etiology of the disease and, thus, leads to ineffective prevention and control of the infectious origin.

Table 6: Serological studies of blood serums of cows and calves

Farm	Animals Groups	Antibody titers of paired serum samples (M±m)			
		Infectious Rhinotracheitis, log		Parainfluenza, lg	
		First study	Second study	First study	Second study
MTF_1	1	0.91±0.28	0.66±0.19	0.75±0.12	0.85±0.12
	2	0.58±0.19	0.58±0.19	0.70±0.18	0.56±0.18
MTF_2	1	0.08±0.08	0.25±0.19	0.56±0.18	0.67±0.18
	2	0.25±0.19	0.41±0.19	1.09±0.22	1.20±0.22
MTF_3	1	––	––	1.01±0.22	0.82±0.22

	2	––	––	1.11±0.22	0.93±0.20
MTF_4	1	0.17±0.17	––	0.75±0.18	0.90±0.20
	2	––	––	1.01±0.12	1.01±0.12
MTF_5	1	0.25±0.19	0.25±0.19	0.80±0.20	0.64±0.20
	2	0.33±0.08	0.50±0.19	1.04±0.20	1.20±0.20
MTF_6	1	––	––	1.35±0.22	1.17±0.12
	2	––	––	1.22±0.22	1.11±0.22
MTF_7	1	––	––	1.11±0.22	0.93±0.20
	2	0.08±0.08	0.25±0.19	1.17±0.12	1.17±0.12
MTF_8	1	––	––	0.93±0.12	0.82±0.21
	2	––	––	1.04±0.20	1.22±0.22
MTF_9	1	0.50±0.19	0.33±0.19	1.25±0.22	1.06±0.12
	2	0.41±0.19	0.25±0.19	1.45±0.22	1.27±0.22
MTF_{10}	1	––	––	0.93±0.22	0.77±0.22
	2	––	––	1.22±0.22	1.04±0.20
MTF_{11}	1	––	––	1.04±0.20	1.22±0.22
	2	––	––	1.17±0.22	1.17±0.22
MTF_{12}	1	––	––	0.67±0.18	0.56±0.18
	2	––	––	0.59±0.18	0.45±0.18

In experimental farms, there were also no positive cases of detection of viral diseases of animals (TABLE 6). In all farms where we conducted research, recorded endometritis (from 14.2 to 27.6%), mastitis (10,3-12,5%) in cows, and gastrointestinal (16,8-50,2%), respiratory (7,6-10,5%) diseases of newborn calves. Most often, Purulent-catarrhal endometritis (67.5%), acute catarrhal (69.2%) and acute serous (22.7%) mastitis, as well as enteric form of gastrointestinal diseases (67.3%) and catarrhal-purulent bronchopneumonia (81.2%) in newborn calves were most often registered. Studies have found that mastitis and endometritis in cows, as well as gastrointestinal and respiratory diseases in newborn calves are caused by an almost similar Association of microorganisms that circulate in the farm (intra-farm infection).

Thus, recently, in the field of veterinary infectious pathology, phenomena have been observed that make it necessary to revise old concepts. Previously, infectious diseases with a complex etiology were preferred to be considered according to the principle "disease = macroorganism + microorganism". This was convenient when researchers dealt with pathogens of high pathogenicity. But this concept turned out to be untenable with associated infections, which have become widespread recently.

This situation indicates significant changes that have occurred in the world of microorganisms. At the same time, under the influence of environmental factors, unsatisfactory food supply, violations of basic veterinary and sanitary rules for keeping and milking, failure to fully implement measures to prevent infectious diseases and break the epidemiological chain, the immunocompetent system of the macroorganism changes, immunodeficiency occurs, and the protection of the animal's body weakens. In such conditions, not only virulent, but also conditionally pathogenic, secundary microflora and even saprophytes can be activated. This situation creates wide opportunities for the formation of various combinations of parasitic agents in various biotopes of the animal organism and the occurrence of parasitocenoses.

CONCLUSION

In this way, our detailed analysis of the epidemiological situation with regard to cattle diseases caused by conditionally pathogenic bacteria, shows that in farms of the Moscow region, which are unfavorable for factor infections, there are all conditions for the circulation of pathogens, multiple passages through susceptible animals, an increase in virulence of conditionally pathogens, and in some cases the emergence of pathogenicity in saprophytic bacteria.

The results of the research show that the associations of conditionally pathogenic microorganisms which circulate among cattle in farms of the Moscow region, represent microbiocenoses which in livestock biotope are independently regulated under the influence of various factors of animal's organism and external environment. Evolutionary-ecologically created parasitocenoses are an integral part of a strictly balanced complex ecosystem of biogeocenoses of farms, each of which affiliates is an important component in the etiology of the development of factor infections cattle's disease. Analysis of the species composition of isolated microorganisms shows that cow's endometritis and mastitis, as well as newborn claves's gastrointestinal and respiratory diseases are caused by conditionally pathogenic microorganisms, which, in most cases, are common representatives of the intestinal tract and biotopes of skin and mucous membrane of healthy young animals. However, with a decrease in immunobiological resistance of the body and unfavorable environmental conditions, these saprophytic microorganisms may cause or complicate the infectious process.

Obtained data show that the composition of microbiocenoses of farms in the Moscow region is subject to seasonal and age-related fluctuations, and also largely depends on sanitary-hygienic and zootechnical feeding conditions and keeping animals. Thus, the analysis of biocenosis of 12 farms in Moscow region, which are

unfavorable in terms of obstetric and gynecological diseases of cows, as well as gastrointestinal and respiratory newborn calves's diseases shows that the main factors that contribute to the development and dissemination of these pathologies are unsatisfactory feed base, violations of elementary veterinary and sanitary rules of maintenance and milking, failure to fully implement measures to prevent these diseases due to the lack of sufficient material base in farms. All this indicates that farms do not have adequate conditions to ensure the epidemiological chain during infectious process.

An effective fight against malignant microbial ecosystems requires in-depth knowledge of the quantitative and species composition of parasitocenoses, study of relationships between its individual representatives, imperative identification of pure culture of all its affiliates, the determination of the marker biological properties of microbial agents, and the detailed biocenotic diagnosis of the animal habitat (farming, barn, etc.).

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CONFLICT OF INTEREST

The authors have no conflict of interest.

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