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Etiopathogenesis, Diagnostics, And Treatment Of Bovine Mastitis

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Abstract

Bovine mastitis is caused by the pathogenic and opportunistic pathogenic microflora affecting the mammary gland.Hematologic studies of cows with mastitis have shown a reduced level of erythrocytes and haemoglobin and a proved increase in the number of leukocytes and erythrocyte sedimentation rate (ESR). It has been found that the concentration of total protein, calcium, phosphorus, carotene, glucose, cholesterol in the blood serum of diseased animals is lower, and the AAT and ALT activity is more intense than in healthy animals.Among the cows, sick with a clinical form of mastitis, the use of Mamicur with local application of ichthyol ointment proved to be the most efficient treatment method.

Key words: Mastitis, diagnostics, milk, anti-mastitis preparations, blood

1. Introduction

Every year, a substantial percentage of milking herd loses its economic value due to reduction or total loss of milk productivity and reproductive ability. Combating bovine mastitis is among the most important problems of dairy cattle breeding. According to the International Dairy Foods Association data, annually 2% of cows suffer from the clinical form of mastitis, and up to 50% from the subclinical form of mastitis [1]. Bovine mastitis inflicts more significant damage on dairy cattle breeding[2,3], than all non-contagious diseases taken together. The morbidity rate of cows in the herd may vary from 10 to 55%, about 75% of livestock therewith may have suffered from the disease [2-5]. Incidence of mastitis increases as the size of herds increases, computer technologies are introduced, and cow productivity improves. Losses involve early cow disposal, failure to produce sufficient milk and calves, expenditures for diagnostics and treatment of sick animals [6,7].

To some extent, mastitides emerge due to such pathologies of reproductive organs as endometritides, subinvolution of uterus, retention of placenta, and others, since in the course of their development different pathogenic microbes and their toxins from the female reproductive organs, degradation products of effusion accumulated in the uterine cavity, enter its mammary gland.

Of importance in mastitis etiology and pathogenesis is a microbial factor, its interaction with the mechanisms of local immune defence of the mammary gland and general immunobiological reactivity of the body, that defines the specifics of the disease manifestation and progression [8,9]. The milk of cows, sick with mastitis, always has pathogenic microorganisms (staphylococci, streptococci, E. coli, and others) with a certain antibiotic resistance, which create a permanent source of infection in the mammary gland. The milk of such animals is neither edible, nor suitable for feeding newborn animals.

Various forms of mastitis should be controlled, based on the current knowledge of etiopathogenesis of this pathology type, and incorporate diagnostic, treatment and prevention stages, with the components adapted at most to the technological level and economic state of the dairy cattle breeding sector.

Endocrine-vascular interrelationships and interactions between the female mammary gland and reproductive organs represent a theoretical basis for developing comprehensive schemes of medical-preventive activities in mastitides and pathologies of animals' reproductive organs. In most cases, treatment of bovine mastitis commences even after the disease clinical signs had appeared. Under production conditions, subclinical mastitis most often becomes clinical in several weeks, hence the therapy measures taken not necessarily produce a beneficial effect.

To date, various plans of comprehensive treatment and prevention of mastitides in cows have been developed and employed, however not always these methods are efficient and flawless

[10-12].The work was aimed at studying etiological aspects of mastitides in cows during lactation period, early diagnosis methods, and selection of optimum treatment regimens for females with the mammary gland pathology.

2. Materials and methods

Scientific and production tests were carried out on the basis of a commercial dairy farm of the "Rodina" JSC in Zhukovsky district, Bryansk region. The farm specialises in commercial dairy production.

Etiological structure of mastitis was analysed in the "Bryansk Interregional Veterinary Laboratory" FSBI through bacteriologic examination of the udder secretion from 48 diseased cows with clinical and latent forms of mastitis. Bacteriologic studies of the udder secretion were conducted using commonly accepted classic methods pursuant to approved instructions. In case of detecting a clinical form of mastitis, an increase in the afflicted udder quarter, tenderness on palpation, raised local temperature were observed; enlarged supramammary lymph glands and changes in the milk quality were registered. Overall condition of an animal was also taken into consideration: suppression, deterioration, or loss of appetite, raised body temperature.

Morphological composition of the whole blood was examined on fifteen 5 year old cows using the Haematology Analyzer Abacus junior vet 5.

In the "Dubrovskaya Regional Veterinary Laboratory" SFI of the Bryansk region, 15 blood serums of black-and-white cows with clinical and latent form of mastitis and clinically healthy were examined for total protein, alkali reserve, carotene, calcium, and phosphorus content. Biochemical blood parameters were analysed according to the "Methodology Instructions on applying standard biochemical methods to examine blood, urine, and milk in veterinary laboratories".

The AAT and ALT activity was defined using the Reitman-Frankel method as modified by B.V. Korovkin. Alkaline phosphatase, alpha amylase, glucose and cholesterol content were found with biochemical analyser BioChem Analette.In farming conditions, mastitis is diagnosed employing clinical examination techniques, sedimentation samples, express-methods applying diagnostic agents. Allowance must be made for the fact that diagnostic agents of various producers have different sensitivity and not always react at the threshold level of somatic cells in milk.

For early recognition of mastitis, 200 cows of various age, yield, and lactation period were examined. The milk from each cow was analysed using diagnostic agents "Masttest", "Ecotest", and "Kenotest", employed to detect somatic cells in milk. Somatic cells in milk are represented by epithelial cells, macrophages, and leucocytes. When the mammary gland is infected, the number of leucocytes in milk increases sharply, what constitutes one of defence mechanisms, since leucocytes perform phagocytosis of bacterial cells [13]. Examinations were made during evening milking operation. After turning the milking plant off, the last milk strips were drawn to the PMK-1 milk-control plate. The results were recorded as per instructions for each preparation.

A sedimentation sample was used for control. In the end of milking, 10ml of milk were selected and placed to the fridge for 16h. Sedimentation sample had a positive result, if there was a deposition and flakelike, viscous, mucous cream.

Laboratory examination of milk was made using Lactoscan MCC and Somatos V 2К analysers.To treat the cows with subclinical mastitis, an experimental and control group of cows were formed (12 animals in each). Females from the first experimental group intramammary received 1 syringe of masti veyxym to the afflicted udder lobe 2 times per day during 3 days and ceftonit subcutaneously at a dose of 1ml per 50kg of animal weight, once a day, during 5 days.The control group cows intramammary received mastiet forte at a dose of 8g 4 times with 12h interval. Mastisept cream was applied locally, daily, every 12h within 2 days.

Cows with clinical mastitis were treated based on etiotropic treatment [14]. 14 cows 3-5 years aged with inflammation processes in the mammary gland tissues were tested. Experimental and control group were formed according to the analogues. The experimental group was treated as follows: mamicur intramammary at a dose of 10ml (1 injector) three times with 12h interval and ichthyol 10% ointment 2 times per day, the control group was treated as per the conventional scheme, accepted in this farm (daily one-time intracisternal introduction of tetramast – 5ml with 4 days therapy course; local application of 10% ichthyol ointment 2 times per day with a thin layer onto the afflicted udder quarter).All digital material was mathematically processed with inference of the Student’s significance.

3. Results and discussion

Bovine mastitis represents the most common pathology, different private farms and dairy farms are confronted with. It is dangerous since it can strike an animal in any season and physiological state: during drying-off, lactation, after calving [15]. The microbial factor contributes to the development of the mammary gland inflammation, because in most cases pathogenic microflora is given off from secretion of the lobes afflicted with mastitis in bacteriological examination [16].

When diagnosing for mastitis, bacteriological examinations are mandatory since they characterise reliably the mammary gland contamination and affection rate. About 85% of all bovine mastitis cases are due to the microbial factor. Staphylococci and streptococci, which either cause diseases themselves, or overlap the pathological process, caused by other factors, are principally given off from the secretion of afflicted udder quarters. According to Firsov G.M. [17], it has been found that 36.47% of tested cultures were classified as staphylococci, 26.30% of cultures were categorised as streptococci; 2.20% - as E. coli, 11.1% - as mixed forms of microorganisms (staphylococci, streptococci, and Escherichia coli in various combinations). At that, no microflora was detected in 23.93% of samples.

To identify the main contagious matters for defining the etiological structure of bovine mastitis, there has been carried out a bacteriological examination of secretion from the udder with lobes afflicted with mastitis (Table 1).

Table 1. Species composition of microflora of the udder secretion of cows, sick with mastitis

Derived microflora	Clinical mastitis, n=24		Subclinical mastitis, n=24	
	Cultures derived		Cultures derived	
	number	%	number	%
Staphylococcus aureus	15	21.7	9	14.8
Staphylococcus epidermidis	3	4.3	8	13.1
Staphylococcus haemolyticus	7	10.1	4	6.6
Streptococcus agalactiae	12	17.5	9	14.8
Streptococcus dysgalactiae	5	7.3	5	8.2
Micrococcus	2	2.9	6	9.8
Enterococcus faecalis	11	15.9	8	13.1
Escherichia coli	6	8.8	7	11.4
Pseudomonas aeruginosa	2	2.9	2	3.3
Candida	3	4.3	2	3.3
Aspergillus fumigatus	3	4.3	1	1.6
Total	69	100	61	100

Analysis of the data of microbiological and mycological examination of the species composition of the udder secretion microflora of cows, sick with clinical and subclinical mastitis, has revealed that 130 cultures of microorganisms, with 9 species of microfungi among them, were derived from 48 milk samples. Derived cultures in clinical progression of mastitis in 79.7% of cases are represented by gram-positive cocci, in 11.7% – representatives of the Enterobacteriaceae and Ps. aeruginosa family, and 8.6% - candida fungi and a causative agent of aspergillosis - Aspergillus fumigatus. In the latent form of mastitis, gram-positive cocci were also predominant – 80.4% with only 19.6% percentage of other pathogenic microflora representatives. Other researchers mention that there are many strains of pathogenic gram-positive cocci in the milk of dairy cows. Zverev E.V. [17] in his studies states that in the milk of cows, sick with mastitis, coccigenic microflora is detected in 90.0% of cases, in 65.4% represented by staphylococci, in 13.9% - by streptococci, and in 16.7% of cases by mixed microflora. Other scientists [18,19] indicated that coccigenic microflora has a predominant role in etiology of mastitides, sorting it out in 57.1% of cases.

Mostly monocultures (Staphylococcus aureus, Staphylococcus haemolyticus Staphylococcus epidermidis, Streptococcus agalactiae, Streptococcus dysgalactiae, Enterococcus faecalis, E. coli, Micrococcus) are sorted out in bacteriological examinations of the udder secretion of cows, sick with the clinical form of mastitis. Among mixed representatives the following associations were detected: Staphylococcus aureus + Enterococcus faecalis, Staphylococcus aureus + E. coli + Enterococcus faecalis, Staphylococcus aureus + E. coli + Enterococcus faecalis + Candida, E. coli + Enterococcus faecalis.

In subclinical mastitis, monocultures of coccigenic microflora were identified, which had less intense virulent activity. From 24 milk samples, in 14.8% of cases Staphylococcus aureus was separated; in 13.1% - Staphylococcus epidermidis; in 6.6% - Staphylococcus haemolyticus. Of streptococci, Streptococcus agalactiae (14.8%) and Streptococcus dysgalactiae (8.2%) species were detected. Percentage of associations while examining secretion from the udder with latent mastitis was observed far less frequently.

A clinical form of mastitis is seen with low body and udder resistance, when a causative agent reproduces, and at the same time its pathogenicity strengthens; it occurs more often in unsatisfactory housing conditions.

When there are inflammation processes in the mammary gland, blood serves as an informative marker [20]. It fulfils various functions and ensures vital activity of all body tissues. When physiological state of the animal body is normal, the peripheral blood composition and properties are more or less constant [21].

Analysis of haematological indicators from Table 2 shows that the blood indicators of diseased cows, suffering from clinical mastitis, are subject to considerable changes. On the part of white blood, it is seen that the number of leucocytes increases twofold, as compared to clinically healthy animals ($P<0.01$). It indicates a prominent leucocytosis, related to the development of the exudative phase of inflammation. A reduction in the concentration oferythrocytes in the peripheral blood of sick cows is accountable to the fact that the inflammation process in the mammary gland has a suppressive effect on erythrogenesis [22]. Blood haemoglobin saturation decreases by 30.0% (P<0.05). The value of haematocrit has also reduced by 20.7%. Of note were substantial changes on the part of ESR, its growth therewith occurred with statistically significant difference in indicators, as compared to clinically healthy animals (P<0.05). Under normal conditions, erythrocytes are charged negatively on their surface, hence, their sedimentation rate is not high. During inflammation processes, the content of coarse proteins in the blood plasma increases. They, settling on the surface of erythrocytes, reduce their negative charge, what leads to their coming closer together and increases the sedimentation rate.

In the second group of animals with the latent form of mastitis, changes in the mammary gland were less prominent. Thus, count of erythrocytes was higher by 7.35%, ESR value - by 26.57%, number of leucocytes – by 34.68% as compared to control analogues.

Table 2. Haematological indicators of cows, sick with mastitis (n=15)

Indicator	Normal range	Group of animals		
		I experimental (clinical mastitis)	II experimental (subclinical mastitis)	Control (healthy)
Erythrocytes, 10^{12}/l	5-7.5	4.73±0.23	7.15±0.6	6.66±0.19
Haemoglobin, g/l	90-130	76.4±5.1*	104.2±4.6	109.2±4.9
Haematocrit, %	35-45	28.8±1.27	32.06±0.97	36.3±1.28
ESR, mm/h	0.5-1.5	1.85±0.05*	1.05±0.04	0.75±0.06
Leucocytes, 10^9/l	4.5-12	16.7± 0.34**	12.6±0.63*	8.23±0.37
Banded neutrophiles, %	2-5	9.53±0.05**	5.14±0.56	3.8±0.15
Segmented neutrophils, %	20-35	54.2±2.84*	33.4±2.8	30.9±1.16
Eosinophiles, %	3-8	10.47±1.2*	6.33±2.2	3.58±0.46
Monocytes, %	2-7	6.8±0.34	4.2±1.17	2.6±0.41
Lymphocytes, %	40-65	18.68±2.27**	50.77±4.6	59.1±1.8
Basophils, %	0-2	0.32±0.12	0.16±0.08	0.02±0.02

Note: *P<0.05; **P<0.01 as compared to clinically healthy animals.

The leukogram analysis has revealed that the total number of banded neutrophiles and segmented neutrophils in cows with the clinical form of mastitis is definitely higher as compared to clinically healthy animals by 60.1 and 42.9%, respectively. Neutrophils are considered the first line of defence against bacteria causing mastitis, and often they are targeted when developing treatment and prevention technologies [23,24]. Pronounced neutrophilia in sick animals indicates the inflammation process in the mammary gland and translocation of neutrophils along the blood channel into the site of inflammation. In addition, an actual 3.2 times reduction in the number of lymphocytes has been observed in sick cows. An increased number of eosinophiles in blood is most commonly associated with allergy reactions in the body in response to the presence of pathogenic microorganisms in the mammary gland and intensification of the exudative phase of inflammation [23,24].

Changes in the leukogram of cows with latent mastitis were less pronounced. Only the quantity of banded neutrophiles was larger than the standard values for a physiologically normal state, as compared to control analogues by 26.1%.

The data of Table 3 show that while the pathological process develops in the mammary gland, some indicators of biochemical status of cows have changed substantially.

Table 3. Biochemical indicators of cows, sick with mastitis, (n=15)

Indicator	Normal range	Group of animals		
		I experimental (clinical mastitis)	II experimental (subclinical mastitis)	Control (healthy)
Carotene, µmol/l	0.40-1.0	0.21±0.08**	0.15±0.03**	0.72±0.07
Calcium, mmol/l	2.5-3.1	1.9±0.12*	2.65±0.23	2.9±0.28
Phosphorus, mmol/l	1.4-1.9	1.1±0.12	1.6±0.13	1.81±0.25
Alkali reserve, volume % CO_2	45-65	55.2±3.25	61.7±2.24	57.7±4.16
Total protein, g/l	72-86	56.7± 2.34*	62.6±0.63*	81.24±2.45
Glucose, mmol/l	2.22-3.33	1.09±0.06**	1.84±0.19*	3.12±0.12
Cholesterol, mmol/l	2.1-5.0	1.2±0.08*	2.4±0.18	3.9±1.23
ALT activity, u./l	25-50	104.7±7.2**	76.27±2.25*	43.64±3.42
AAT activity, u./l	30-90	106.9±2.37	94.2±2.49	72.7±2.41
Alkaline phosphatase activity, u./l	40-65	78.25±1.76	62.87±4.36	59.6±1.8
Alpha amylase activity, u./l	41–98	42.71±2.14	85.26±4.08	78.3±3.09

Note: *P<0.05; **P<0.01 as compared to clinically healthy animals.

Carotene is the main source of vitamin A in the body of animals. It enters the body with food and depends on conditions of cropping forage cultures [25-27]. The uptake of carotene occurs in the intestines, the liver is the repository of vitamin A. Malnutrition, even with sufficient supply of carotene, facilitates 1.5-2 times decrease in the synthesis of vitamin A [28]. A proved reduction in the carotene content 3.4 and 4.8 times, respectively, has been observed in the blood serum of examined cows with the clinical and latent forms of mastitis. In spring, we noted a low value of this indicator, what coincides with calving and is due to the increased percentage of low-carotene concentrate in the diet of animals [29].

Biochemical studies of blood characterise rather fully the metabolism state in the body. Indicators of the content of total calcium and inorganic phosphorus in the blood serum are employed to evaluate whether the mineral nutrition is balanced or not during various lactation phases. Calcium is absorbed in the form of complex compounds in the small intestine. It decreases excitability of muscle and nervous systems. In the body of animals, its major portion is related to phosphoric acid, being the basis of the bone tissue [30].

Phosphorus exchange is closely related to calcium metabolism. Optimum ratio between calcium and phosphorus in forages is 2:1, which can be achieved using a mineral fertiliser [31-33]. Lactation period affects the calcium-phosphorus metabolism. In the period of high milk yield, cows are unable to uptake as much calcium and phosphorus from forage, as they release them with milk, hence, they utilise these elements from the bone tissue.

When exploring the amount of calcium and phosphorus, it was found that cows with the clinical form of mastitis have a tendency towards reduction in these indicators relative to control by 34.5 and 39.2%, respectively. It indicates that with this pathology, the body utilises these elements from the bone tissue. In animals suffering from the subclinical form of mastitis, the indicators of the content of total calcium and inorganic phosphorus were not beyond the limits of a physiologically normal state.

Buffer systems of blood serve an important role in controlling the acid-alkali balance and body balance control of animals. The alkali reserve rate in experimental groups was within the range of standard values of a physiologically normal state. It can be stated that no state of acidosis develops in the body of cows, what is obvious from the alkali reserve rate, corresponding to standard values [34].

Blood proteins in the body of animals fulfil numerous functions, namely: maintain constant pH of blood, oncotic pressure, level of cations in the blood, and play an important role in forming immunity. In case the amount of proteins entering the body is in deficiency, caused by their insufficient amount in the forage when using mineral nutrition improperly, a retardation of growth and development is noted, productivity decreases [35-37].

Concentration of total protein in the blood serum of experimental animals, sick with the clinical andsubclinical form of mastitis indicates the low level of protein nutrition. The indicator was lower than the values in the control group by 30.2 and 22.9%, respectively.

The nature of carbohydrate and fat metabolism in the body of animals can be inferred by the concentration of glucose and cholesterol in the blood serum, and the activity of alpha amylase enzyme.

Concentration of glucose, the primary precursor of lactose, in the blood is the principal indicator of carbohydrate metabolism. Glucose is absorbed into the blood and enters tissues, its excess is deposited in the liver in the form of glycogen, in muscles, and other tissues [38]. From the results of examining the glucose level in the blood of cows, sick with mastitis, hypoglycaemia was noted, the changes therewith were significantly different (first experimental group – 1.09±0.06; second experimental – 1.84±0.19; control – 3.12±0.12). Lack of glucose causes gluconeogenesis when the body uses protein and fat resources for energy needs.

Cholesterol is an important structural element of the cell membrane, involved in the formation of complexes with proteins. It is needed for biosynthesis of biliary acids, corticosteroids, estrogens, and androgens [39]. A proved 3.3 times reduction of cholesterol in the group of animals with clinical mastitis indicates a pronounced stress-response, since pathological process in the mammary gland causes a diseased reaction.

Alpha-amylase is an enzyme, actively involved in the digestion process. In cows with the clinical form of mastitis, it was near the lower limit of a physiologically normal state.

Aminotransferases (aspartate and alanine aminotransferase) are involved in the reactions of enzymatic transfer of aminogroups between aminoacids. They are found in all organs and tissues of animals, but the maximum activity is seen in the liver, skeletal muscles, myocardium. Aspartate and alanine aminotransferases in the liver transform aspartate and alanine to keto-acids - oxaloacetic and keto-acetic, which are used in the gluconeogenesis reactions for synthesis of glucose and glycogen [40].

The ALT and AAT activity throughout the experiment had a tendency towards growth in experimental groups, but in the first group its activity was more essential as compared to the second one by 27.1 and 13.5%, respectively. The activity of aminotransferases in sick cows increases since their bodies started to actively use proteins for energy needs.

Inflammation in the mammary gland in cows is manifested by some complex signs: reduction in productivity, increase in local temperature, augmentation and induration of quarters, etc. Colostral milk and milk obtained from the cows with mastitis, reduces the immunobiological reactivity of calves, causes retardation of growth, and even death of young stock [41].

Laboratory diagnostics enables detection of sick animals. The clinical state of the udder quarters [42, 43] can be promptly inferred from various diagnostic studies of the bovine mammary gland secretion using dimastine, mastidine, kenotest, masttest.

Under production conditions, the dependence of the mammary gland disease on age, productivity and lactation period was examined. It has been found that the maximum amount of females with the latent form of mastitis is observed in the end of lactation, as well as in cows more than 7 years old. The morbidity rate in this age group amounted to 3.5% of all the livestock.

To detect bovine mastitis, express-diagnostic agents were employed that allow diagnostics of the pathology development at early stages. Among the examined cows, 18 animals with the latent form of mastitis were detected. Diagnostics was performed based on sedimentation sample, with determination of the number of somatic cells by the "Somatos V 2K" analyser. At that, the use of diagnostic agent "Kenotest" enabled identification of 17 sick animals, "Masttest" – 12 and "Ecotest" – 7, what demonstrated 94.4, 66.6 and 38.9%, respectively, diagnostic efficiency of used preparations.

Digital data show that physical and chemical indicators change in mastitides (Table 4). Of note is the reduction of fat in cows with the subclinical and clinical forms of mastitis by 23.6 and 33.1%, respectively, relative to healthy animals. The content of non-fat milk solid (NFMS) and lactose in cows with the clinical form of mastitis decreased by 11.03 and 9.6%, respectively, as compared to the control group of cows. In females, suffering from the subclinical form of mastitis, these indicators were almost the same as those for clinically healthy animals.

Milk protein in experimental groups 1 and 2 declined by 20.7 and 4.9% as compared to the control group, while milk acidity increased by 7.1 and 2.7%, respectively, what is explained by cleavage of proteins to ammonia, and sodium bicarbonate entering from the blood [44].

Table 4. Physical and chemical indicators of milk of cows with clinical and subclinical mastitis forms

Indicator	Group of animals, n=5		
	I experimental (clinical mastitis)	II experimental (subclinical mastitis)	Control (clinically healthy)
Fat, %	2.12±0.14*	2.42±0.13	3.17±0.29
NFMS, %	7.6±0.41	8.51±0.08	8.64±0.32
Density, kg/m^3	1026.25±1.73	1029.35±0.54	1030.6±1.85
Lactose, %	4.12±0.43	4.58±0.07	4.56±0.22

Salts, %	0.65±0.05	0.74±0.02	0.76±0.04
Acidity, °T	17.65 ± 0,10	16.85 ± 0.04	16.40 ± 0.04
Protein %	2.6±0.17	3.12±0.05	3.28±0.21
pH	7.35±0.09*	6.92±0.05	6.75±0.12
Conductivity, mS/cm	7.63±0.27**	5.96±0.25*	4.78±0.09
Somatic cells, thsd./cm^3	>1500±0.0**	674±13.6*	108±8.96

*P<0.05; **P<0.001 – difference is statistically significant.

Of practical importance is determination of the milk electrical conductivity. A regularity exists that, with occurrence of subclinical mastitis, more sodium and chlorine ions enter the milk from the blood, hence, the milk conductivity rises. In our experiments, conductivity was reliably higher in cows with clinical and subclinical mastitis, by 37.3 and 19.8%, respectively.However, of maximum practical value in performing diagnostic studies of mastitis is the identification of somatic cells. High concentration of somatic cells is a sign of dysgalactia or a disease.

When examining the milk of animals from the first and second experimental groups, an average amount of somatic cells was >1500 and 674 thsd./ml, respectively, what can be interpreted as a positive and doubtful reaction. In cows from the control group, the number of somatic cells was within standard limits for a physiologically normal state and amounted to 108 thsd/ml.

Due to high incidence of bovine mastitis, of importance is combating this disease. To detect pathology with inflammatory processes in the mammary gland, early diagnostics, prompt efficient treatment and effective prevention have been employed. To date, abundant diagnostic aids, complex anti-mastitis preparations and prevention methods have existed on the veterinary market, however the issue of combating mastitis is still an open question [45].

It should be noted that many applied anti-mastitis preparations, which mostly contain antibiotics, sulfanilamides, nitrofurans, together with desired effects may have some negative side effects on the condition of animals [46-49].

After comparing the obtained results of subclinical mastitis treatment in cows, it has been found that therapeutic efficiency of the complex treatment regimen in the experimental group was a little higher than in the control group (Table 5). Productivity of cows prior to disease was 28.7±1.18 kg/day on an average. Throughout the entire period of disease, of note was a reduction in the milk productivity by 62% on an average. Daily milk yield in this period was about 11kg/day. An increase in the milk yield by 40.2% was observed by the 6^{th} day of treatment, a recovery of the milk productivity was seen 10-12 days after the end of treatment. In the control group, indicators of productivity and somatic cells restored more slowly.

Reduction of the number of somatic cells in the milk of diseased animals serves as an important indicator of the efficiency of provided treatment. 6 days after the end of treatment, there were less than 250 thsd./ml somatic cells in the milk of 62% of cows, and up to 500 thsd./ml in 15.7%. For two cows with the clinical and latent forms of mastitis, the applied treatment regimen had no therapeutic effect, there were more than 1500 somatic cells in these animals.

Table 5. Somatic cells and productivity of cows, sick with latent mastitis, during treatment

Experimental group (n=12)					
Indicator	27.08.19 (1st treatment day)	29.08.19 (3rd treatment day)	31.08.19 (5th treatment day)	06.09.19 (6 days after treatment)	12.09.19 (12 days after treatment)
Daily milk yield, kg	11.18±0.72	11.63±0.65	14.4±1.13	18.69±1.95	22.61±2.77
Number of somatic cells in milk, thsd./cm^3	>1500	1315±37.4	868±22.3	503.3±27.2	–
Control group (n=12)					
Daily milk yield, kg	11.35±0.75	11.41±0.65	13.2±1.15	16.53±1.77	21.88±2.38
Number of somatic cells in milk, thsd./cm^3	>1500	1418±45.4	965±27.6	623.9±21.8	–

When exploring the efficacy of treatment regimens for cows, sick with mastitis, it has been found that therapeutic effectiveness of using mamicur proved to be higher as compared to tetramast, and with mastitis it amounted to 91.7%, at that 92.5% of the udder quarters were cured. Employing tetramast facilitated recovery of 86.2% of cows, sick with the clinical form of mastitis, 87.3% of the udder quarters therewith were cured. Therapeutic manipulations for animals until their clinical recovery were used during 6.2 days with tetramast and 4.8 days with mamicur on an average.

In recent times, veterinary-sanitary and technology-related requirements, imposed on milk, have become stricter. The milk, in which an elevated level of radionuclides – caesium and strontium was identified, is not subject to sale [49]. In conditions of the "Dubrovskaya Regional Veterinary

Laboratory" SFI of the Bryansk region, raw bovine milk (dated 14.06.2019, the sample registration number – 11.06.1010) was examined for caesium 137 as per GOST 32161-2013 and strontium 90 as per GOST 32163-2013 using universal radiometer-spectrometer RSU-01 Signal-M. The permissible level of these radionuclides was below the values of the RD[1] indicator. No inhibiting and antimicrobial agents were detected in the milk as well.

Creation of high-quality products depends on the defects of forage origin. They emerge when animals eat plants with specific odour and taste. Proper selection of forage diets, reduction in doses of forage with odour, exact feeding regimens make it possible to fully avoid feed flavours in milk [50].

4. Conclusion

Quality and quantity of the obtained milk depend on the health condition of a cow, and, particularly, on the condition of its mammary gland, exposed to different diseases. Mastitis is a very common pathology, and the incidence of bovine mastitis in herds may reach 50–60%, what leads to considerable economic losses of dairy cattle breeding sector farms.

Various microorganisms are the main reason for the development of inflammation process in the udder parenchyma. In healthy females, there are no microorganisms in the mammary gland parenchyma, however, they enter the udder galactogenically when defence mechanisms of the cow body are suppressed, causing various inflammatory responses.

Representatives of pathogenic and opportunistic pathogenic microflora with staphylo- and streptococci (80.4%) predominating are the most frequent causative agents of mastitis in cows at the commercial dairy farm of the "Rodina" JSC. In conditions of producing dairy products, pathogens have their effect in association with other causative agents. The percentage of associations while examining secretion from the udder of cows with the latent form of mastitis is less than that of monocultures.

Morphological and biochemical indicators of dairy cows' blood enable evaluation of the state of metabolic processes in the body and its resistance. It should be noted that these indicators deviate from the physiologically normal state according to the degree of inflammation process occurring in the mammary gland. In a strongly pronounced form of mastitis, there have been observed a considerable change in the number of leucocytes (twofold), a proved increase in ESR, a decreased number of erythrocytes, a reduced haematocrit value and haemoglobin content as compared to clinically healthy animals. In animals with latent form of mastitis, these changes in the body were less prominent.

[1]RD – regulatory documentation

In analysing the leucogram, an increase in the number of eosinophiles, banded and segmented neutrophils has been identified in contrast to a considerable proved 3.2 times decrease in the number of lymphocytes.

Biochemical examination of the blood of cows, sick with the clinical form of mastitis, has established that the concentration of total protein, calcium, phosphorus, carotene, glucose, cholesterol proved to be lower, and the ALT and AAT activity – more intense than in healthy animals.

Numerous efficient methods to diagnose the mammary gland dysfunction have been used nowadays in veterinary practice, what makes it possible to assess the form and stage of the inflammation process in the gland alone. Fast mastitis test with a large set of diagnostic agents: 2 and 10% Mastidine solutions, 5% Dimastidine solution, masttest, kenotest, and others have been widely used in production conditions.

In our studies, Kenotest had a maximum diagnostic efficiency (94.4%). During laboratory examinations of milk, characteristic traits will involve: a rise in the number of somatic cells to over 500 thsd./ml, milk PH raised to 7.0–7.5; increased electrical conductivity of milk, a reduction in the amount of protein, fat, non-fat milk solids, and lactose.

To stop an inflammation process in the mammary gland of lactating cows, it is imperative that the efficient anti-mastitis preparations be used, which cause attenuation of the inflammation process in the udder and strengthen the body defence mechanisms.

The proposed treatment regimen (Masti Veyxym+Ceftonit) allowed 83% of cows, sick with subclinical mastitis, to recover.

For cows with the clinical type of mastitis, the use of Mamicur with local application of ichthyol ointment proved to be the most efficient treatment method.

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