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Dissemination, Etiology, Pathogenesis And Treatment Of Cattle Teat Diseases In Agricultural Organizations Of The Sverdlovsk Region Of Russian Federation.

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ABSTRACT

In the highly productive herds of the Sverdlovsk region of Russian Federation, the cattle teat diseases are widespread and are registered by 11.9-56.6% of all examined udder quarters. The spread of teat diseases increases, correspondingly to the growth of milk production. At the same time, a significant percentage is revealed even with the use of automatic milking directly on the udder quarter. The studies showed that there is milking systems influence on the udder condition - the thermographic inspection showed that the udder surface temperature during milking decreases, but the teat surface temperature increases. However, the temperature increase depends on condition and type of milking equipment and teat cup liner. The permanent udder injury by automatic milking is confirmed by the presence of occult blood in 26.2-68.6% of samples. The main etiological factor in the development of teat diseases is the automatic milking, as well as a violation of milking technology and inadequate milking equipment. The location and size of teats, the shape of the udder, and the internal anatomical features of the structure also influence the occurrence of teat pathology and their injury level. In the course of the research, it was noted that if the mucous membrane has many folds in the teat part of gland cistern, then the teat injury level increases. A study of the defected teat internal structure under hyperkeratosis has shown that 35-40% of the length of the teat canal is damaged and shorter by 11.5% compared to nipples having an adequate physiological response to automatic milking represented by a circular callus, as well as an increase in its lumen in the region of the external opening. When the defect is in the form of a circular callus, there is a change in the mucosa by 12% of the length of the canal. Histological examination revealed changes in the teat tissues that characterize post-traumatic state: the presence of ruptures in the mucous membrane, edema, vascular reaction, micronecrosis foci, thickening of the stratum corneum and its sloughing. The use of silicone glycerogyrogel «Silativit» and pharmacological compositions based on it for the treatment of lactating cows with various forms of hyperkeratosis and teat injuries is highly effective and does not affect milking technology; it is evidenced by ultrasound and histological examination of the udder teat tissues. In addition, this drug allows short course treatments, since they have a pronounced prolonged effect on the tissue, despite the persistent effect of the etiologic factor.

Keywords: cattle, productivity, mammary gland, hyperkeratosis, milking, mastitis, organosilicon compounds.

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INTRODUCTION

Cattle teat diseases are a widespread problem in herds with high milk productivity [5, 16, 17]. Due to the fact that they are a predisposing factor in the development of mastitis, an important task is the development of a system for diagnosis, treatment and prevention of these pathologies [7, 11, 19]. On large dairy farms using different types of milking systems, hyperkeratosis is the most common disease of the udder teat [3, 20]. This disease has a polyetiological nature, which is based on the imperfection of milking equipment and features of the anatomical structure of the udder [1, 12, 18].

Purpose and objectives of the study

The purpose and objectives of the study were to assess the spread of cattle teat diseases at enterprises of the Sverdlovsk Region, identify main etiological factors, pathogenesis of the diseases, and develop effective methods for treating these pathologies.

MATERIALS AND METHODS

Analysis of distribution and structure of cattle teat pathologies using the diagnostic scale was conducted in years 2006-2017 in 16 agricultural enterprises of the region with different levels of milk productivity and machine milking technologies. The total number of examined animals was 3567 during this period.

Udder thermograms were performed before and immediately after milking with the «Irtis - 2000 CH» device based on a tripod at a given distance from the udder (0.8-2.5 meters), scanning time was about 3.2 seconds.

Ultrasound examination of the cattle udders was carried out in the interval between milking, so that the cavity of the teat part in the milk cistern was filled with milk. The structure of the glandular part of the milk cistern was studied using an «Ecoson 900V» ultrasound scanner with a linear 7.5 / 6.5 / 5 MHz multifrequency sensor and a water buffer.

The peculiarities of the structure of the milk cistern were studied on the slaughter material obtained from 12 cows with different teat injury level by hyperkeratosis. Milk cisterns were opened with a parasagittal section passing along the axis of the teat canal. After that, several aspects were evaluated: the folds of the mucous membrane of the milk cistern, their location along the circumference of the cistern and the Furstenberg rosette, the presence and nature of the relationship were evaluated. Then, the material was selected for histological examination.

The tissue was handled according to conventional histological techniques. The samples were stained with hematoxylin and eosin, according to Mallory and Weigert. Microscopy and microphotography of histological samples were performed using a microscope «Micros MCX 300» and a video attachment «VIDI-cam».

The research method of residual milk was performed to detect the occult blood. Sampling was carried out immediately after the milking. 15 ml of milk was collected in a test tube. The study was conducted within an hour after collection of the samples. Blood was tested by the centrifugal method. 8-10 ml of milk, heated to 40-45 °C, was poured into a test tube, it was capped and centrifuged for 10 minutes at 1000 rpm. The result was assessed visually by the presence of a reddish sediment at the bottom of the tube.

Direct counting of somatic cells in milk samples from each cow was carried out in the selection laboratory of the Regional Information and Selection Center (RISC) of «Uralplemetcenter» using a combined system of Bentley Instruments (USA), consisting of an infrared analyzer «Bentley 2000» and «Somacount 500» - the somatic cell counter. In the conditions of agricultural enterprises, the number of somatic cells in the samples was determined using a portable optical somatic cell counter of the DeLaval company.

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Clinical studies of pharmacological compositions were carried out in the basic farms of the Sverdlovsk region, with the productivity of cows over 7000 kg of milk, forming groups according to the principle of analogies.

The received quantitative indicators and digital photos are processed on a PC Pentium. Statistical processing of the data was carried out using the computer program Microsoft Excel 2010.

RESULTS OF THE RESEARCH

Studies of available literature sources have shown that known teat pathology classifications represent diseases by the etiologic factor [17]. These classifications do not include the whole range of clinical manifestations of pathology, and they cannot estimate the teat injury level. In connection with this, a new diagnostic scale of teat injury was developed, taking into account the teat state system, by J.S. Britt and R. Farnsworth (1996).

The diagnostic scale, which was proposed by Elisin A.V. and Barkova A.S. (2006), is a panel of 18 photographs that visually reflect the 6 main types of teat injuries: 1 - a slight thickening of the epidermis; 2 - prominent circular callus with insignificant roughness; 3 - rough circular callus with signs of hyperkeratosis and obstruction of the teat canal; 4 - rough circular callus with radial cracks and gaping of the teat canal; 5 - papillomas; 6 - trauma. Photographs are arranged in 3 rows (A, B, C), which corresponds to light, moderate and severe severity (Fig. 1).

Fig 1: Diagnostic scale of cattle teat injuries

The use of this scale in the conditions of agricultural organizations made it possible to identify not only the disease, but also the severity of the process [5].

When analyzing the udder teat conditions of 3567 cows in farms with different levels of milk productivity and milking technologies, it was established that pronounced pathological changes in the area of the canal sphincter are recorded in 11.9-56.6% of all examined udder quarters. The damage in the form of hyperkeratosis complicated by radial cracks and gaping of the teat canal has about 9.3% of the examined cows, and the damage in the form of uncomplicated hyperkeratosis is at the level of 18.7%. At the same time, we found that the most severe types of pathology are recorded in animals in highly productive herds.

The results showed the need to identify the main risk factors of the disease development. Farms were divided into 4 groups, taking into account their productivity level: 3001-4000 kg; 4001-5000 kg; 5001-6000 kg of milk; more than 7001 kg of milk.

In the first group with productivity 3001-4000 kg of milk per year, a loss by a circular rough callus with teat canal obstruction (hyperkeratosis) reported by 12.5% of the investigated teats with hyperkeratosis, radial cracks and teat canal dehiscence (complicated by hyperkeratosis) is marked on 4.3% of the nipples.

In the second group of farms with productivity 4001-5000 kg dairy cows, hyperkeratosis registered in 19.0% teats and complicated hyperkeratosis was 7.0% of all injuries.

In the third group of enterprises, with milk productivity at 5001-6000 kg, damage in the form of hyperkeratosis was noted in 8.5% of all cases, and in the form of complicated hyperkeratosis - in 3.2% of all cases.

In the fourth group of farms, with the milk productivity over 7000 kg, was a significant increase in the number of severe forms of teat lesions. Pathology in the form of complicated hyperkeratosis was 16.6%. Regular hyperkeratosis was in 21.3% of all injuries.

A significant influence is provided by the technology used for keeping and milking animals, as well as the technical condition of the milking system and teat cup liner. However, with an increase of milk production of more than 9000 kg per cow, even the use of robotic milking does not prevent the development of hyperkeratosis in teats of varying severity [9]. So, with the productivity of 10000 kg of milk per cow and robotic milking, the level of hyperkeratosis was 30%, and complicated hyperkeratosis - 14.4%.

To determine the effect of machine milking on the udder state, the udder thermography was performed before and after milking. The effect of machine milking on the temperature of the surface of the udder's teats was studied in two agricultural organizations. In one farm milking was carried out using a linear milking machine «ADM-8»,the other farm used milking system «Europarallel» by DeLaval company.

Comparison of the systems according to the peculiarities of their influence on the temperature of the udder during the milking process was carried out under normal operating conditions. Cows without udder pathologies were tested by thermography.

Thermography was performed before milking (and before udder treatment) and immediately after milking with the «Irtis-2000 SN» device. At the same time, the temperature was tested at three points: in the teat base, the middle and the apex.

At the first dairy complex, we obtained and analyzed 82 thermograms, at the second - 78 thermograms.

The results of measurements were presented as a temperature difference before and after milking. Since the measurements were carried out under the same ambient temperature conditions, their changes did not affect the absolute temperature value, so it was not separately evaluated. Examples of thermograms (photos) are in Fig. 2 (A, B); the images show temperatures at three points: in the teat base, the middle and the apex.

Based on these results, it can be concluded that the temperature of the udder surface during milking decreases, but temperature of the teat surface increases. These results are consistent with the studies of foreign authors [8].

Table 1: Temperature difference at the beginning and the ending of milking at different teat points

Milking system	n	Temperature difference before and after milking		
		Teat base	The middle	The apex
ADM-8	82	0,42±0,044*	1,75±0,031*	2,26±0,036*
Europarallel	78	1,12±0,026	0,80±0,024	1,28±0,032

*– the difference is reliable, P<0,05

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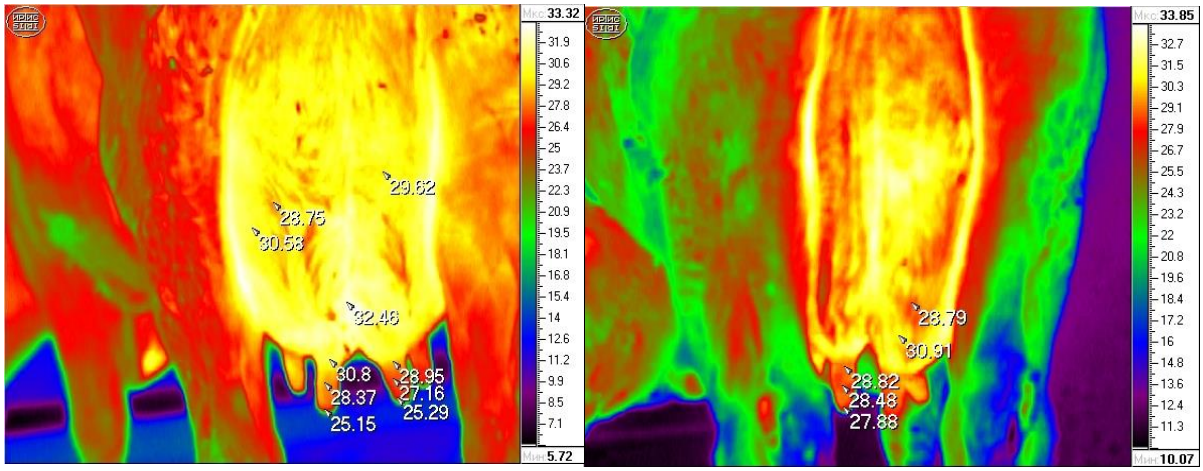
When milking with «ADM-8», the temperature increase was found at all points. The average temperature in the apex, when this system is used, increased by 5.9% -8.7% (1.8-2.9 °C).



A B

Figure 2: Examples of udder thermograms

When milking with «Europarallel», the temperature in the apex was less pronounced and reached only 2.8% -6.0% (0.9-1.9 °C). In this case, the average temperature of the teat base was lower by 1.12 °C, compared to the results before milking.

This allows us to conclude that there are differences between different milking systems in relation to their influence on the teat temperature, and the thermography method allows to predict the temperature before and after milking.

To determine the presence of permanent udder injury caused by the milking system, 67 samples of residual milk were examined. The presence of occult blood was found in 26.2% of all samples of robotic milking and 68.6% of linear milking system (the manufacturer was practically irrelevant).

The next stage was to study the anatomical features of the udder structure in order to determine the factors that predispose the disease development.

To determine the correlation between the udder structure features and the frequency of teat damage, a morphometric evaluation of udders was performed.

Correlation between the distance between the anterior and posterior teats and their injuries was not established. However, the distance between the anterior and posterior teats affected the occurrence of hyperkeratosis. With a distance less than 10 cm, the level of hyperkeratosis was at 59%, and the complicated form of hyperkeratosis was noted at 37.6%. At a distance of 10-12 cm serious injuries at the apex were noted at 57%, and at a distance of more than 10 cm - by 38.5% of the cases.

When the distance between the posterior teats is less than 8 cm, the level of hyperkeratosis was at 53.8%, while with the distance between the posterior teats of 8-10 cm – is at 24.1%.

The shape of the teat apex has an influence on the morbidity as well. With a flat and rounded shape the level of hyperkeratosis was at 60%, with a pointed apex – at 78.6% of all teat injuries, of which 70.8% was a complicated form of hyperkeratosis.

It was noted that animals with the capacious soft udder have more severe forms of teat diseases. The number of lesions in these cows is at the level of 70.6%.

The relationship between the teat wall structure, the mucous membrane of the milk cistern and the predisposition to teat damage is traced.

Teat ultrasonography of 67 lactating cows showed that cows that did not have any pathological changes of the teat canal aperture had a short teat canal and also a thin teat wall structure. In the presence of circular callus and an uncomplicated form of hyperkeratosis, the thickness of the teat wall was about 6 mm, and the length of the teat canal was ca. 14 mm. If there was a lesion at the teat apex in the form of complicated hyperkeratosis, there was also a tendency to shorten the length of the teat canal, which in this case we associate with partial destruction of the tissues of the teat canal [2].

Table 2: Morphological characteristics of teats in various forms of hyperkeratosis

Types of lesions	n	Measurements	
		Length of teat canal, in mm	Thickness of teat wall, in mm
Slight thickening of the epidermis	45	12,7±0,24	5,4±0,12
Circular callus	64	14,2±0,28	6,0±0,13
Hyperkeratosis	42	14,1±0,27	5,9±0,15
Complicated hyperkeratosis	15	12,9±0,76	5,3±0,15

Ultrasound showed the presence of profound changes in the tissues of the teat canal, which make up 17 to 30% of its length. With complicated hyperkeratosis, almost 4.5 mm of the teat canal is damaged, which is about 35% of its length, which is 2 or more times higher than in canals with lighter forms of the injury.

In severe clinical forms of teat injuries with hyperkeratosis, tissues at the apex have significant lesions (Fig. 3). The ultrasound picture marks the changes in the teat canal, even its configuration changes. It also has an increased echogenicity due to thickening of the mucous membrane, and its funnel-shaped extension is noted in the external aperture, which can be 2 times its thickness.

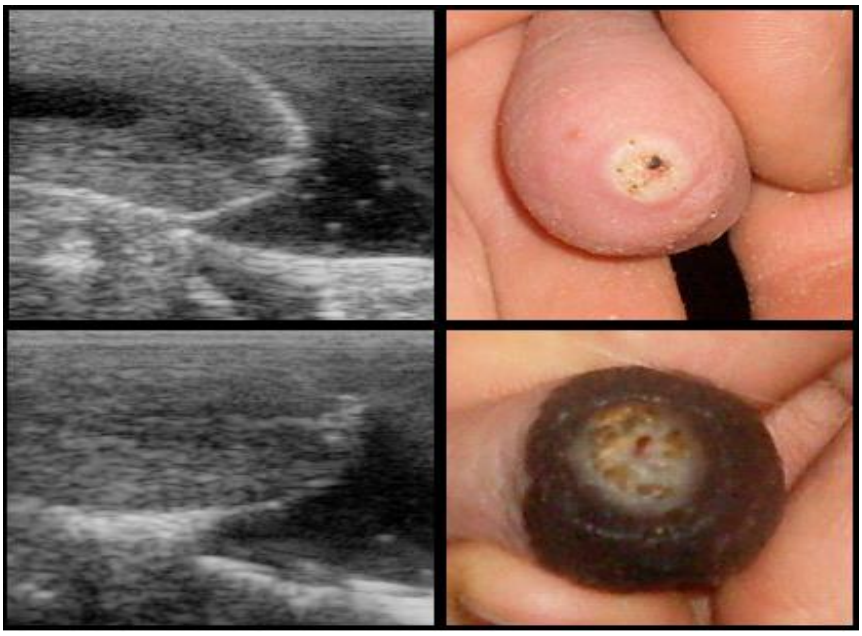


Figure 3: Ultrasound and clinical pictures of uncomplicated form of teat hyperkeratosis (upper row - anterior right teat; lower row - posterior right teat)

In severe cases of hyperkeratosis, there is a thickening, hardening and an outwards inversion of the mucous membrane of the teat canal.

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The next stage was to study how does the folds of the milk cistern mucous membrane affects the teat damage level. Ultrasound was performed between milking, so that the cavity of the milk cistern was filled with milk.

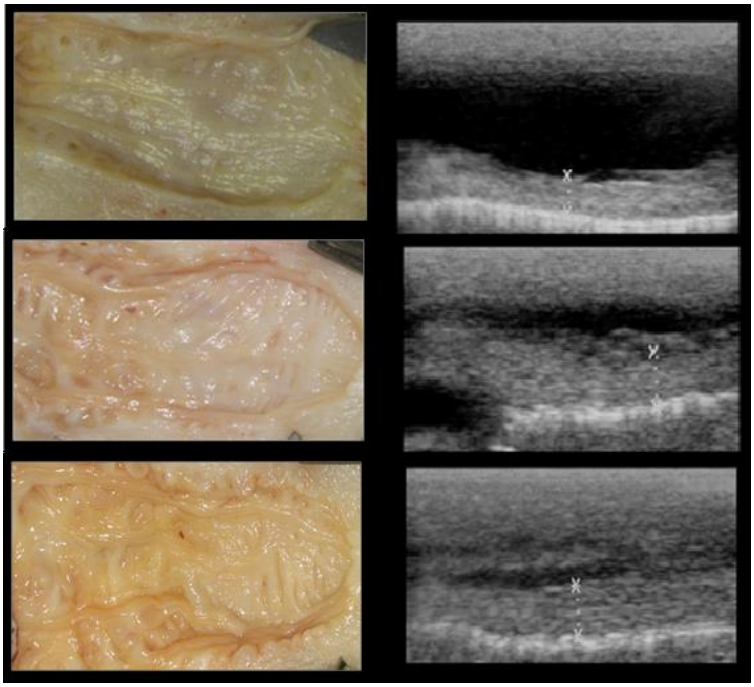

Figure 4: Macroscopic and ultrasound pictures of the folds of the milk cistern mucous membrane. From top to bottom: folds are absent, moderate folding, vast folding of the mucous membrane

The study of the longitudinal folds intensity showed that 6% of teat folds were as a solid array; 21% had an expressed folded mucous membrane; at 45% the folding was moderate; and at 28% the folding was almost not expressed at all (Figure 4). This research showed that if the level of mucous membrane folding increases - then the teat injury becomes worse. In the presence of complicated hyperkeratosis the longitudinal folding was significantly expressed in 53% of all cases, and with circular callus - 22%, but the folding in a form of solid array has not been identified.

The most frequently recorded level of mucous membrane folding is moderate, which was noted on average by 40-50% of all cases with each kind of injury.

The study of teat internal structure during hyperkeratosis revealed a damage of 35-40% of the length of the teat canal and it's shortening by 11.5% in comparison with teats that have an adequate physiological response to machine milking represented by a circular callus, lumen in the area of the external aperture. The length of the mucous membrane was shortening by 12%, because of circular callus.

In the second and third stages of the pathological process, the surface of the callus becomes rough, individual keratin plates begin to peel off. This leads to accumulation of pathogenic bacteria in the keratin layer and causes contamination of the pathological process of radial cracks formed at the third stage, which prevents their healing. This creates mastitis pathogens formations in the proximity of the teat canal [6, 15, 21].

With the complication by the infectious process of radial cracks, a further development of the disease is noted, which can take a severe form, accompanied by functional and structural changes in teat tissues [12].

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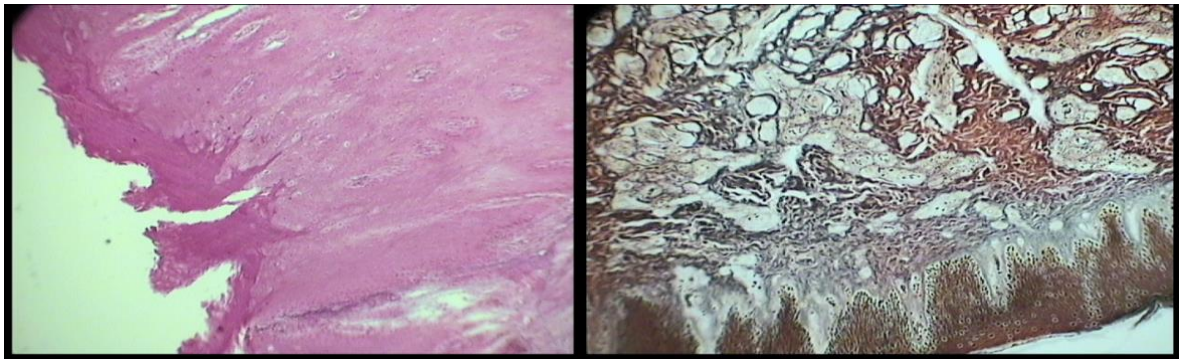
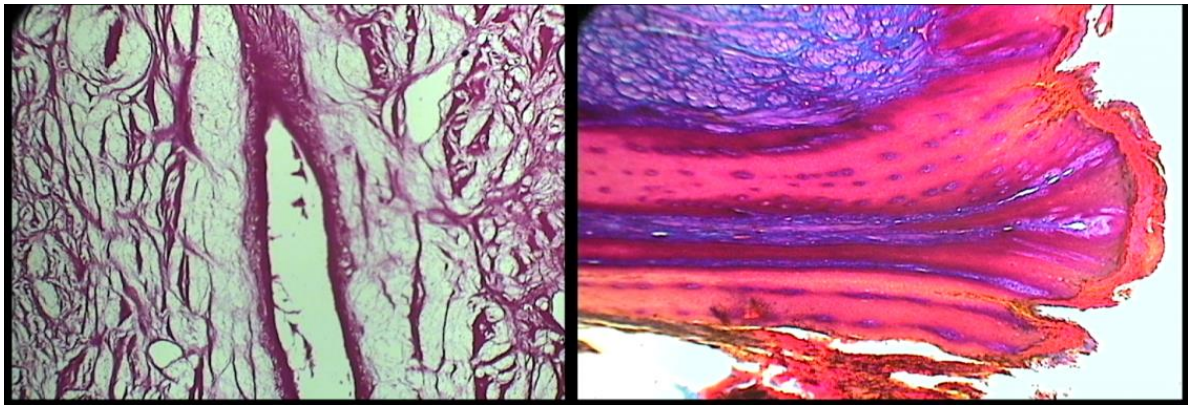


A B

Figure 5 A: Mucosal ruptures in the external apertures of the teat canal, Weigert stain, 40x. B: Zone of edema - enlargement of lymphatic capillaries, silver impregnation, 100x



A B

Figure 6 A: Perivascular edema in the teat apex, Weigert stain, 100x. B: Degradation of elastic fibers in the outer part of the teat canal. Rupture of mucous membrane in the region of the teat canal aperture; epithelium cornification, the thickening of the stratum corneum and its sloughing; micronecrosis along the skin rim around teat canal, Mallory stain, 40x

Histological examination revealed changes in teat tissues, characteristic for the post-traumatic state: the presence of ruptures in mucous membrane, edema, vascular reaction, micronecrosis foci, thickening of the stratum corneum and its sloughing (Fig. 5, 6).

The purpose of the studies was to elucidate the relationship between cattle teat lesions with hyperkeratosis and the level of somatic cells in the milk. Therefore, we have conducted milk analysis from 353 cows to detect somatic cells (SC) in it.

Depending on the level of SC animals were divided into three groups. The first group - the SC containing in milk is up to 200 thousand/ml (considered as a healthy mammary gland); the second group - the SC containing in milk is 201-500 thousand/ml (infection or irritation of the udder); the third group - the SC containing in milk is more than 500 thousand/ml (latent mastitis). When cows were distributed according to the degree of hyperkeratosis, there was paid attention to the most obvious pathological process on the teat.

Analysis indicates that in animals with less than 200 thousand/ml of SC in the milk, the complicated hyperkeratosis is registered in 11% of all cases; animals with SC containing ranging from 200 to 500 thousand/ml in milk are up to15%, and in cows with the number of SC in samples more than 500 thousand/ml - increases up to 18%.

That information allows us to conclude that the teat hyperkeratosis is a risk factor in the development of infection and inflammation of the udder. This is confirmed by the results of the study, where we analyzed the state of the udder and teats in cows with an increased content of somatic cells in milk.

At the same time, the presence of these lesions does not always lead to an increase of SC in the milk, as the natural resistance of the animal's organism, the number and pathogenicity of microorganisms and other factors also influence the development of the inflammatory process in the tissues of the mammary gland [9, 21].

The next stage of our study was to develop methods for the treatment and prevention of hyperkeratosis in highly productive cows during lactation. Together with the Institute of Organic Synthesis named after I. Ya. Postovsky of Ural Department of Russian Academy of Sciences we have developed means for topical application on the basis of biologically active compounds of silicon, which possess anti-inflammatory, wound healing, regenerating and transcutaneous activities [4]. Silicone agents intensify the biosynthesis of collagen, protect tissues from drying and edema, they are not toxic, they do not accumulate or do not cause local and systemic allergic reactions [10, 14].

We studied the organosilicon glycerohydrogel, which was developed in the Institute of Organic Synthesis named after I. Ya. Postovsky of Ural Department of RAS (the patent of the Russian Federation № 2255939, C07F7 / 04, which is published 10.07.05, the Bulletin №19).

Chemically, it is a (2,3- dioxipropil)-ortho-silicate glycerohydrogel (formula 1), which is the basis substance of «Silativit».

$$Si(C3H7O3)4 \cdot xC3H8O3 \cdot yH2O, \qquad (1)$$

where $3 \le x \le 10$; $20 \le y \le 40$.

«Silativit» is a colorless or slightly yellow substance, with an oily consistency, odorless, with a wide range of viscosity.

Clinical study of the efficacy of pure «Silativit» and several compositions based on it has been conducted by treating cattle teat hyperkeratosis under conditions of industrial milk production. Evaluation of the results was made with the diagnostic scale of teat injuries.

The study of the «Silativit» effectiveness during hyperkeratosis was performed on 24 cows, divided according to the principle of analogue pairs into the experimental and control groups, with 12 cows in each.

During the treatment course, after application of «Silativit», the teats' skin became soft and elastic in the experimental group, softening of the circular callus in the area of the teat canal sphincter was noted, and the layers of keratinized cells were eliminated. A decrease by 3.9 times (as compared to the initial information) in the number of teats with complicated radial cracks in the form of hyperkeratosis was found. A significant improvement with granulation of cracks and leveling of the surface of the teat apex was noted in 27.1% of the cases.

Under the influence of this medicine, severe forms of lesions gradually change into lighter ones until they are completely cured. Under the influence of mechanical factors, the formed scab came off, the height and diameter of the callus are reduced, and the injury was considered only thickening of the epidermis according to the scale.

The study of cows 7 days after the end of treatment showed further improvement in the state of the teat apex tissues. The restoration of the skin pattern was noted. In the experimental group, by that time, 29.2% of teats had no pathological changes. Serious forms of lesions were noted at 39.5% of all teats; the complicated hyperkeratosis was noted only by 2%.

In the study, which was 14 days after the end of treatment, was found that the number of teats without pathological changes continued to increase (by 1.3 times) and the number of severe nipple lesions decreased by 1.5 times compared with the results of the study on the 7th day after the end of treatment.

The study of cows 21 days after the end of the course of therapy showed that there was a further increase in the number of teats that did not have any pathological changes (by 1.1 times). This indicator was at

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39.6%. However, there was an increase in the number of teats with pathology in the form of uncomplicated hyperkeratosis by 1.2 times (at 33.4%).

This result can be more clearly assessed by the dynamics of severe forms of teat lesions by hyperkeratosis during the experimental period presented in Fig. 7.

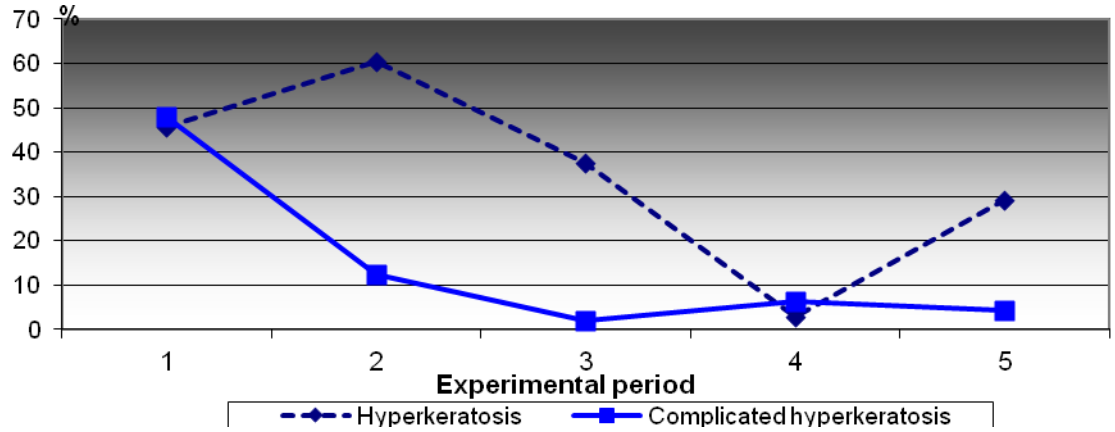

Figure 7: Dynamics of severe forms of teat hyperkeratosis (1 - before the treatment; 2 - after the end of treatment; 3 - after 7 days; 4 - after 14 days; 5 - after 21 days)

Quantitative characteristics of callus diameter in the animals of the experimental group showed that after the treatment, the callus diameter decreased by 3.2 mm. With further measurement, there was a gradual decrease in the diameter of the circular thickening; 21 days the end of treatment, the thickening was at 3.6 mm.

The next stage was to study the therapeutic efficacy of the antimicrobial composition based on «Silativit», containing 0.5% metronidazole, 1% pefloxacin, 0.05% chlorhexidine bigluconate and the «Silativit» as the rest. Due to the fact that teat cracks are a formation of pathogenic and opportunistic microorganisms and constitute a risk factor for udder infection, the number of somatic cells in the milk of each udder quarter was taken into account when forming the groups. The counting of somatic cells in milk was made using an optical counter DCC by DeLaval. Animals with clinically distinct mastitis were not included in the experiment.

Two groups were formed according to the principle of analogue pairs with 22 cows in each. The composition was applied as a thin layer on the teats of animals of experimental group immediately after milking, twice a day for 7 days. No treatment was performed in the control group.

The results of studying the dynamics of teat lesions during the entire experimental period with the use of the antimicrobial composition based on «Silativit» are presented in Table 3.

Table 3: Efficacy of antimicrobial composition based on «Silativit» in the treatment of teat hyperkeratosis

Study duration from the beginning of the treatment	Level of lesion, in %			
	Slight thickening of the epidermis	Circular callus	Hyperkeratosis	Complicated hyperkeratosis
Initial data	0	2,3	18,2	79,5
7 days	11,4	23,8	36,4	13,1
14 days	13,1	23,8	50	13,1
21 days	14,3	40,5	39,3	5,9
28 days	41,7	34,5	23,8	0
35 days	11,9	46,4	29,8	11,9

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The study of animals of the experimental group showed that there was an improvement in the state of the teat apex tissues within three weeks after the end of treatment. The rejection of cornified epithelial cells in the teat canal aperture in a form of dense, rough crust was noted.

The results given in Table 3 also indicate that by this time the number of somatic cells in the milk of the experimental group was significantly decreased in comparison with the initial indices.

Table 4: The number of somatic cells in the milk when using an antimicrobial composition based on «Silativit»

Group	n	Daily milk yield, in kg	Number of somatic cells, in thousand/ml	
			Initial data	The 14th day
Experimental	22	21,4±1,6	274,8±22,3	162,6±14,8*
Control	22	19,8±2,1	286,4±22,3	312,4±32,3

*– the difference with the control group is reliable, P<0,05

Dynamics of severe forms of teat hyperkeratosis during the experimental period when using an antimicrobial composition based on «Silativit» is shown in Fig. 8.

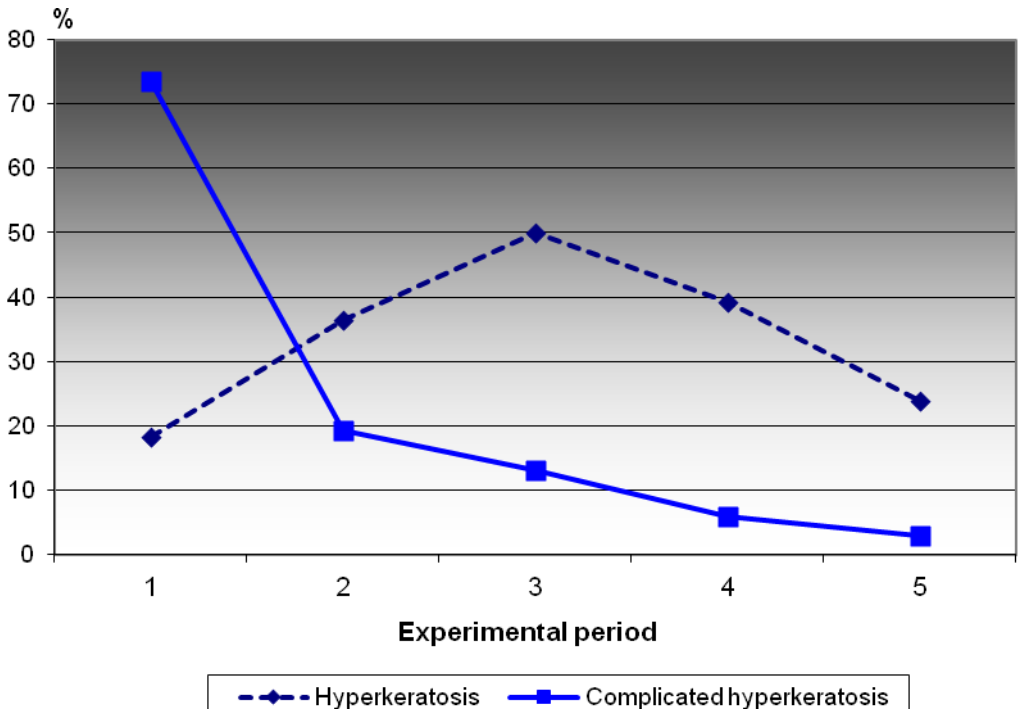

Figure 8: Dynamics of severe teat hyperkeratosis (1 - before treatment; 2 - after treatment; 3 - 7 days after completion of treatment; 4 - 14 days after completion of treatment; 5 - 21 days after completion of treatment)

The presented graphs show the same trends in the action of the antimicrobial composition as in the use of «Silativit» without pharmacological additives: rapid regeneration of cracks in the sphincter region and prolonged aftereffect.

The measurement of the diameter of the circular callus showed that during the entire experimental period, the diameter of the circular callus gradually decreased.

The third stage of research on silicone-based compounds was the study of an ecologically pure pharmaceutical composition containing herbal anti-inflammatory components. The composition uses

supercritical carbon dioxide extract of calendula. According to its physical properties, the composition is a translucent yellow gel, with a characteristic smell of calendula.

The ratio of components, mass. in %: extract of calendula - 0.5; «Silativit» - the rest.

To conduct an experimental clinical study on the principle of analogs, two groups of cows of 14 in each with complicated teat hyperkeratosis were selected. When forming the groups, the number of somatic cells in milk was taken into account. Counting was carried out with the help of a somatic cells counter of DCC milk from DeLaval. Animals with clinically pronounced mastitis were not included in the experiment.

Cows of the experimental group were given 0.5 g of gel per teat immediately after milking 2 times a day for 14 days. Control group animals were treated with gel «Anikol», according to the procedure described above [8].

Evaluation of the results was carried out using the diagnostic scale of teat injuries. To study the regenerative processes in the teat, the ultrasound method was used. Changes in the regime of maintenance and exploitation of animals have not been carried out.

When assessing teats condition before the start of the treatment, all animals of the experimental and control groups had similar lesions. The percent of udder quarters with an excess of 500.000 somatic cells per ml of milk was 33.93% in the experimental group, and 32.14% in the control group.

As a result of the study, it was found that healing of cracks occurred on the 7th day of the treatment course in 96.4%; improvement during the pathological process with the rejection of keratinous layers on the teat apex was noted as well.

In the control group by this time the number of teats with hyperkeratosis complicated by cracks decreased by 1.37 times compared to the initial indices.

In the study of cows of the experimental group 7 days after the treatment course, a further reduction of severe forms of teat lesions was noted, which gradually passed into lighter ones until they were completely cured.

If we consider teat fractures as a separate nosological form, then it can be concluded that the pharmaceutical composition based on «Silativit» containing 0.5% of the CO_2 extract of calendula has 100% therapeutic efficacy on this pathology, the average healing time for teat cracks was 5.1 day. Therapeutic efficacy of «Anikol» was 83.9%, the average duration of tissue regeneration in the area of cracks was 9.4 days, which was significantly longer than in the experimental group (Table 5).

Table 5: The effectiveness of the composition with a CO_2 extract of calendula on the basis of «Silativit» on the skin cracks in the teat apex

Index	Composition with a CO_2 extract of calendula on the basis of «Silativit»	«Anikol»
Therapy effectiveness, %	100	83,93
Crack healing period, in days	5,1±0,4*	9,4±1,2

*– the difference with the control group is reliable, P<0,05

Observations of the animals during the next two weeks showed further improvement in the state of the tissues of the teat apex in the cows of the experimental group. By the fourteenth day after the treatment course, uncomplicated hyperkeratosis accounted for 30.4% in the experimental group. The control group also registered a positive dynamics in the teats state.

21 days after the end of treatment, the positive effect of treatment remained in the experimental group, while in the control group the number of teats with hyperkeratosis increased significantly (up to 55.4%). In addition, the number of teats with complicated hyperkeratosis increased by 2 times (up to 14.3%).

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Measurement of callus diameter in the animals of the experimental group showed that the average diameter at the end of the observation period decreased by 3.1 mm, in the control group by 1.9 mm.

Decrease in the level and severity of teat lesions naturally led to a decrease in the content of somatic cells in milk (Table 6). By the end of the observation period, the somatic cell level was at 256.400 cells per ml milk in the experimental group and it was significantly lower in comparison with the control group. At the same time, the number of udder quarters with somatic cells more than 500.000 thousand per ml of milk decreased in both groups and amounted to 19.0% in the experimental group, and 26.8% in the control group.

Table 6: Somatic cells quantity in the cow milk

Quantity of somatic cell, in thousand/ml	Control group	Experimental group
Initial indices	538,6±22,3	567,8±62,3
14 days after the treatment course	437,6±72,2	256,4±34,4*

*– the difference with the control group is reliable, P<0,05

The obtained results by comparing two medications for topical application showed a higher efficacy of the composition with a Calendula extract based on «Silativit» in the case of teat hyperkeratosis complicated by cracks in highly productive cows.

To assess the change in the state of the teat canal, when the composition, containing 0.5% of CO_2 Calendula extract, was applied, an ultrasound examination of the teats was carried out, during which the composition was applied for 7 days (right quarter of the udder). Evaluation was carried out before the application, after the treatment, and also one week after the end of treatment.

The ultrasonic picture after the application of the composition was characterized by a gradual decrease of the teat canal lesion area, which was visualized on the echogram as a normalization of the echogenicity of this area after 7 days of medicine application (Fig. 9). Seven days after the end of the application, further improvement of the condition of the teat canal tissues was noted, which was manifested by a decrease in the length of the teat canal with increased echogenicity of the tissues. This data obtained by the method of ultrasonography can confirm the prolonged effect of the composition.

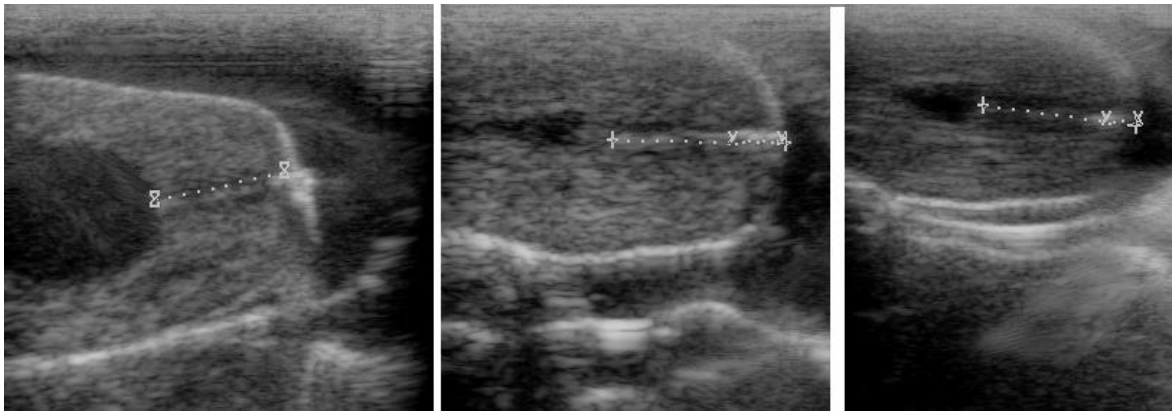

Figure 9: Ultrasonic evaluation of the teat canal condition after the composition application: A – before the treatment; B - after the treatment; B - a week after the end of treatment

To assess the effect of the pharmaceutical composition on the teat skin, histological examination of the tissues of the teat canal aperture was performed. There were rejected cells of the stratum corneum (keratinous callus cells) after application of the composition on the teat apex, which are represented on the histological sample by disparate stratum masses. In the cells of the epidermis granular layer, signs of dystrophy are noted: cytoplasm vaucolisation, expressed karyorrhexis (Fig. 10). Keratin callus is rapidly rejected, due to the replacement of dying and dead cells with new ones. Expressed changes in the basal layer of the skin were not established.

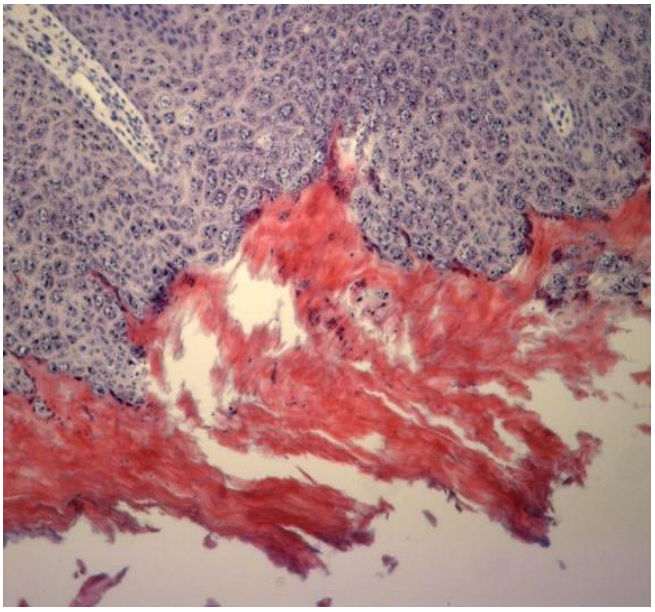

Figure 10: Changes in the epidermis after application of the composition of CO_2 extract of calendula and «Silativit», hematoxylin and eosin stain, 150x

The layered callosity was removed and keratinization processes was inhibited, which contributed to the restoration of the morphological structure of the epidermis.

CONCLUSION

The proposed diagnostic scale of teat injuries in the conditions of agricultural enterprises allows correctly identifying the type and level of teat damage and determining the effectiveness of therapeutic and preventive measures.

In the highly productive herds of the Sverdlovsk region of the Russian Federation, the teat diseases in cows are widespread and are registered by 11.9-56.6% of all the examined udder quarters. The level of milk production, the location and dimensions of teats, and the birth defects of the teat canal sphincter affect the appearance of pathologies and their level.

The influence of milking systems on the udder condition was studied by the method of thermography and showed that the temperature of the udder surface during milking decreases, but the temperature of the teat surface increases. However, the level of temperature increase depends on the condition and type of milking equipment and teat cup liner. The permanent trauma of udder with robotic milking is confirmed by the presence of occult blood in the milk in 26.2-68.6% of all samples.

The level of milk production, the location and size of teats and the shape of the udder affect the appearance of teat pathology and the level of damage [12, 13]. The study of teats anatomical features by ultrasound scanning showed that the main reason for the development of hyperkeratosis is the errors in the technology of machine milking [16, 17].

In the course of the research, it was noted that if the level of mucous membrane folding of the teat area in the milk cistern increases, then the forms of teat injuries become heavier. Thus, in the presence of complicated hyperkeratosis, longitudinal folding was significantly expressed by 53% of all teats, and in case of circular callus - by 22%, with no continuous folding revealed.

A study of the teat internal structure during hyperkeratosis in teat canal aperture has shown that 35-40% of the length of the teat canal is damaged and its shortening by 11.5% compared to the teats that have an adequate physiological response to robotic milking represented by a circular callus, as well as an increase of

the aperture lumen. In the case of circular callus, we also established a change in the mucosa by 12% of the length of the teat canal.

Histological examination revealed changes in the teat tissues that characterize the post-traumatic state: presence of mucosal ruptures, edema, vascular reaction, micronecrosis foci, thickening of the stratum corneum and its sloughing [9].

The use of silicone glycerogyrogel «Silativit» and pharmacological compositions based on it as the treatment of lactating cows with various forms of hyperkeratosis and teat injuries is highly effective and does not affect milking technology, as evidenced by ultrasound and histological examination of the udder and teat tissues. In addition, this medication allows short course treatments, since it has a pronounced prolonged effect on the tissue, despite the persistent effect of the etiologic factor.

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