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Combined anti-Mastitis Composition

Abstract

For the first time, studies were conducted to learn the therapeutic efficacy of tissue-drug therapy using a tissue preparation and a mixture of naphthalene oil, olive oil, camphor oil, analgin and streptocide powder. The traditional method of antibiotic therapy was used as a comparison. Comparing the results of the therapeutic efficacy of the administered drugs showed that the method of tissue-drug therapy is more effective, since when using antibacterial drugs, milk obtained from dairy cows is allowed to be used for food purposes only four days after the last case of drug use, and slaughter of cattle for meat after seven days, and when treating the method we proposed, using a tissue preparation with a cutaneous method of applying a mixture of naphthalene oil, olive oil, camphor oil, analgin and streptocide powder to the udder area, milk obtained from dairy cows is allowed to be used for food purposes immediately after recovery.

Keywords: *Tissue preparations, naphthalene oil, camphor oil, analgin, streptocide, udder lobes, mastitis, antibiotics, streptococci*

Introduction

According to the European Association of Animal Breeders and numerous studies, clinical mastitis is detected in 25% and subclinical mastitis in 50% of dairy cows.

Intensive research is underway to develop and implement safe and highly effective treatments for bovine mastitis.

The etiological factor contributing to the development of mastitis in animals is pathogenic and conditionally pathogenic microflora. Antibiotic-resistant strains are emerging among the isolated strains.

Therefore, there is an urgent need to develop a method for treating mastitis caused by antibiotic-resistant bacterial species.

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Despite antibiotics being the primary therapeutic agents for mastitis treatment, they are associated with serious problems. Thus, to improve the effectiveness of mastitis treatment, there is an urgent need for a new, effective, alternative therapeutic agent.

Research

Tissue biopreparations are increasingly used in the pathology of many infectious diseases, including mastitis (Bodiev, 1983; Pasechnikova, 2007). These preparations have a therapeutic effect in diseases that are difficult to treat with traditional methods. Tissue therapy can also be combined with the use of antibacterial drugs (Voitenko, Kartushina, Pushkareva, Bondareva, 2014).

Tissue therapy stimulates and normalizes various functions of the animal's body, improves the trophic function of the nervous system, increases the function of the thyroid tissue, adrenal glands, pancreas, increases the formation of adrenocorticotropic hormone, the release of corticosteroids, the secretory and motor function of the gastrointestinal tract, gas, intermediate and phosphorus metabolism, and regenerative processes (Vasiliev, 2008; Voitenko, Kartushina, Pushkareva, Bondareva, 2014).

Tissue preparations, unlike antibiotics, lack antimicrobial activity, but their bioactive substances influence metabolic processes in the animal's body, reducing or even stopping inflammation at the site (Sotnikova, 2006; Vasiliev, 2008).

Given the above, the aim was to study tissue-drug therapy to complement and enhance the therapeutic effect.

Materials and methods. The study was conducted at a private farm of Veli Kuliev, Kararkh village, Samukh district, and the regional laboratory. Monitoring the health of the animals, a dispensary examination was performed. The examination revealed several cows with signs of mastitis.

Milk from diseased cows was aseptically collected from udder quarters for bacteriological examination. Teats were treated with 70% alcohol before sampling. Milk samples from each cow were collected in numbered tubes, corresponding to the cow's number. The milk samples were examined in the zonal bacteriological laboratory (Belkin, Cherepakhina, Popkova, Skrebneva, 2010).

First, a reductase test was performed. Six tubes, numbered for each cow, were placed in a rack. 20 ml of milk sample and 1 ml of methylene blue were added to each tube. The mixture was incubated at 37°C. Decolorization time of methylene blue was determined at 20 minutes, after 20 minutes, and after 2 hours. Data are presented in Table 1.

Methylene blue decolorization rate.	Test tube 1	Test tube 2	Test tube 3	Test tube 4	Test tube 5	Test tube 6	Quality assessment of milk
Up to 20 minutes.	+	+	+				very bad.
After 20 minutes to 2 hours.				+	+	+	bad.
Two hours or more.							Satisfactory

As shown in table 1, the first three of six milk samples discolored methylene blue faster, indicating very poor quality. In the remaining tubes, discoloration occurred after 20 minutes, indicating poor quality. This confirmed high bacterial contamination.

Cultures were also performed on solid and liquid nutrient media to investigate for staphylococci, streptococci, and enterococci. Colony morphology was studied on meat-peptone agar. The presence of E. coli and Salmonella was determined on Endo agar, and their differentiation was performed.

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Gas production was tested on Kessler medium. Anaerobes were tested on Kitt-Tarozzi and Wilson-Blair media. After incubation, smears were prepared from colonies grown on nutrient media, Gram-stained, and examined under a microscope. Gram-positive cocci, arranged in pairs or short chains, were identified. Blood agar was used to study the hemolytic properties of the isolated strains, and Kartashova's selective media (liquid and solid) and Edwards medium were used to identify streptococci (Bozhenov, 2007).

Small, dewdrop-like colonies with a zone of hemolysis were observed on blood agar. In Kartashov's broth, the medium changed from blue to green, indicating the presence of streptococci, which ferment lactose, turning the medium green. On Kartashov's agar, small, transparent colonies grew against a blue background, with the surrounding medium turning green. On Edward's medium, streptococci grew as small, grayish-blue colonies. The isolated streptococci were identified using group diagnostic serum in an agglutination reaction. Str. agalactiae and Str. dysagalactiae were identified (Danilevskaya, 2010).

For a tissue preparation from freshly slaughtered cattle under strictly aseptic conditions, parenchymal organs (spleen, liver, and adrenal gland) were taken in the following weight ratio: 89% spleen, 10% liver, and 1% adrenal gland.

After thorough defatting, the spleen, liver, and adrenal gland were refrigerated at 4°C for 6 days. Following this, the organs were minced, and the resulting mince was diluted with isotonic solution at a ratio of 1:2. The mixture was extracted for 2 hours at room temperature with constant stirring. The suspension was then boiled for 1.5 hours and cooled for 3 hours at room temperature. The resulting mass was squeezed through a dense cloth, and the expressed liquid was centrifuged. The supernatant was poured into sterile vials and autoclaved for 1 hour at 1.5 atm. Bacteriological tests were performed to check for sterility (Daricheva, 2000).

Research findings. Clinical examination revealed tenderness, fever, and hyperemia in specific udder areas of 8 cows. Animals showed poor appetite and low milk yield.

For therapeutic purposes, animals were divided into two groups of four animals each. Group 1 animals received a biostimulator subcutaneously in the middle third of the neck at a dose of 1 ml per 10 kg of body weight, and the udder was treated with a mixture of 100 g of naphthalan oil, 100 g of olive oil, 20 g of camphor oil, 10 g of analgin, and 20 g of streptocide powder. Group 2 animals received an intramuscular injection of a penicillin and streptomycin mixture at a dose of 1g/20kg of body weight for therapeutic purposes. Treatment regimen (Polyantsev, 1982).

Daily monitoring of the cows was implemented. Recovery was assessed by changes in general condition, body temperature, swelling, tenderness, and mammary gland secretion.

Key indicators of therapeutic efficacy were reduced disease duration, the dynamics of the disappearance of main clinical symptoms (general condition, temperature response, tenderness of the affected area, etc.), and treatment costs. The rate of decline in main clinical symptoms was assessed starting from the 3rd day after drug administration (Gorbatova, Gunkova, 2009).

Group 1 animals, by day 3 of treatment, showed restored appetite, local temperature, and milk yield.

Group 2 animals, by day 3 of treatment, showed restored appetite, a slight decrease in local udder temperature, and pain responses upon udder palpation.

Experienced animal group	Biostimulant and a mixture of naphthalan oil, camphor oil, analgin, and streptocide powder for topical use.	Penicillin and streptomycin mixture
Group 1.	Subcutaneous biostimulant 1 ml / 10 kg body weight, and the drug mixture applied topically 3 times daily after milking.	
Group 2.		Once daily, 1g/20kg body weight. Intramuscularly

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On day 7 of the treatment course, the overall condition of the animals in the first group improved, appetite returned, udder temperature normalized, and hyperemia disappeared. Animals in the second group also showed improvement, with the temperature of the affected area gradually decreasing. Milk samples were collected from each group for a repeat reductase test (Gorlov, Yurina, Slozhenkina, 2008).

Results of the reductase test are shown in table 3. Methylene blue decolorization rate.	Test tube 1	Test tube 2	Test tube 3	Test tube 4	Test tube 5	Test tube 6	Quality assessment of milk
Up to 20 minutes.							Very bad.
After 20 minutes to 2 hours.							bad.
Two hours or more.	+	+	+	+	+	+	Satisfactory
5 ½ hours and more.							Good.

As shown in Table 3, the decolorization of methylene blue occurred after 2 hours of incubation in a thermostat with a mixture of milk and methylene blue. The bacterial content in the milk, as well as their enzymes, was low, and the milk sample quality is considered satisfactory. Comparing the results of the reductase test before and after treatment, it can be noted that before treatment, the reductase test for three samples was rated as "very poor," and for the rest as "poor," but after treatment, these indicators were low and rated as "satisfactory" (Krus, Shalygina, Volokitina, 2002).

Conclusion

Treatment methods in all animal groups had the same effect on the body of cows with mastitis. However, comparing the therapeutic efficacy of the administered drugs showed that tissue-drug therapy is more effective. When using antibacterial drugs, milk from dairy cows can be used for food purposes only four days after the last administration of the drug, and slaughter of cattle for meat is allowed after seven days. In contrast, with our proposed method, using a tissue preparation with a topical application of a mixture of naphthalan oil, olive oil, camphor oil, analgin, and streptocide powder to the udder area, milk from dairy cows can be used for food purposes immediately after recovery.

Therefore, our recommended cloth-drug therapy is safe and can be widely used in livestock farms for treating mastitis in cows.

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