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CLINICAL CHANGES AND MORPHOBIOCHEMICAL BLOOD ANALYSIS RESULTS IN CALVES INFECTED WITH DIFFERENT DOSES OF STREPTOCOCCUS PNEUMONIAE.

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Annotatsiya: Maqolada *Streptococcus pneumoniae* infeksiyasining turli konsentratsiya eritmalari bilan zararlangan turli yoshdagi buzoqlarning 2, 5 va 7-kunlaridagi klinik o‘zgarishlari, gemotologik tahlillarning me’yoriy ko‘rsatkichlardan farqlari o‘rganilgan va tadqiqot natijalariga statistik ishlov bo‘rilgan.

Аннотация: В статье изучены клинические изменения у телят различного возраста, инфицированных *Streptococcus pneumoniae* растворами различной концентрации, на 2-е, 5-е и 7-е сутки, а также отклонения показателей гематологических исследований от нормативных значений. Результаты исследования подвергнуты статистической обработке.

Abstract: The article investigates the clinical changes observed on days 2, 5, and 7 in calves of different ages infected with various concentrations of *Streptococcus pneumoniae*. Differences between hematological parameters and their normal values were analyzed, and the research results were statistically processed.

Калит сўзлар:

Buzoq, qo‘zg‘atuvchi, zararlash dozasi, konsentratsiya, *Streptococcus pneumoniae*, tana harorati, nafas soni, yurak urushi (puls), gemotologiya, mikrob tana, klinik o‘zgarish, leykotsitar formula, bazofill, eozinofil, neytrofil, laboratoriya.

Key words:

Calves, pathogen, infectious dose, concentration, *Streptococcus pneumoniae*, body temperature, respiratory rate, heart rate (pulse), hematology, microorganism, clinical changes, leukocyte differential count,

basophils, eosinophils, neutrophils, laboratory analysis.

Relevance of the Topic: In the livestock farms of our republic, various infectious diseases are widely among young animals, causing significant economic losses to farms. Diseases such as pasteurellosis, colibacteriosis, salmonellosis, and streptococcosis—often occurring as secondary infections in young animals, especially calves, lambs, rabbits, chicks, and others—are known to be present in almost all regions of the country.

To prevent these diseases, all newborn animals in farms must be vaccinated in a timely manner according to established schedules. At a minimum, diagnostic examinations should be conducted once or twice a year. However, at present, due to the insufficient production of necessary biological preparations for disease prevention within the republic, certain shortcomings are observed in the implementation of epizootic control measures.

Currently, streptococcosis infection is frequently observed among young animals, particularly in calves, where *Streptococcus pneumoniae* often causes acute disease forms in many regions. Taking this into account, it is important to analyze available literature and determine the degree of infection caused by streptococcal agents among young animals—especially calves—as well as to study the clinical course of the disease, its seasonality, and the extent of general pathological changes in the organism.

Researchers emphasize that the spread and coverage of infectious diseases depend not only on their etiology but also on a number of other factors. It has also been noted that it is difficult to establish a strict boundary between infectious and non-infectious diseases in young animals [9; pp. 445–446].

Streptococcosis is an infectious disease of farm animals caused by *Streptococcus pneumoniae*. It is characterized by inflammatory processes in the lungs and intestines, sepsis, arthritis, and severe pathological changes in the lymphatic system.

In young calves, when caused by *Streptococcus pneumoniae*, streptococcosis manifests in peracute, acute, and subacute forms, often leading to high mortality. In older calves and cows, the disease typically follows a chronic course, although acute cases may also occur, leading to conditions such as endometritis, abortion in cows, arthritis, and mastitis.

Streptococci are facultative aerobic bacteria that grow well at +37°C in both liquid and solid nutrient media. In meat-peptone broth, bacterial growth causes slight turbidity. The growth characteristics, colony formation, and coloration of cultures are determined in suitable nutrient media [8; p. 160]. The growth intensity increases when cultured in semi-liquid agar supplemented with 5% defibrinated blood serum.

It is well known that among young animals present in all livestock farms and household sectors of our republic, calves in particular are highly susceptible to various infectious diseases and are prone to infection.

Results of the Study: Experiments were conducted on a total of 9 calves aged from 7 days to 3 months, based on the principle of analogous pairing. In the first experimental group (7–30 days old), *Streptococcus pneumoniae* infection was administered intraperitoneally at a dose of 2 billion microbial cells per 1 ml. In the second group (1–3 months old), a dose of 3 billion microbial cells per 1 ml was administered intraperitoneally.

In the third (control) group, 3 ml of physiological saline solution was injected into the abdominal cavity (Table 1).

Experimental infection of calves using different concentrations of Streptococcus pneumoniae (n = 9). (Table 1)

Groups	Number and age	Name of the pathogen	Route and dosage of infection	Results

	of the calves		Dose	Method	Alive	Died
Group I (Experimental)	n=3,/7-30 daily	*Str.pneumoniae*	2 billion microbial cells per 1 ml	Peritoneal cavity	-	3
Group II (Experimental)	n=3,/1-3 monthly	*Str.pneumoniae*	3 billion microbial cells per 1 ml	Peritoneal cavity	-	3
Group III (Control)	n=3,/1-3 monthly	-	3 ml of 0.9% physiological saline solution	Peritoneal cavity	3	-

Calves in the experimental groups infected with different doses of Streptococcus pneumoniae were monitored for housing conditions, clinical signs, body temperature, heart rate per minute, respiratory rate, and morphobiochemical blood parameters on days 2, 5, and 7 after infection.

In calves of the first and second experimental groups, starting from day 2 post-infection, signs of depression and weakness were observed. Body temperature increased to +40.5–42.4°C, heart rate increased (by approximately 16 beats), and respiration accelerated to 9–11 breaths per minute. The affected calves appeared lethargic, with dry muzzles and hyperemia of the ocular mucous membranes. On days 4 and 5 post-infection, some calves showed decreased appetite or complete loss of appetite. In 7–30-day-old calves, clinical signs such as serous nasal discharge and

labored, wheezing respiration were observed (Figures 1, 2 and Table 2).





Clinical examinations of calves infected with different doses of Streptococcus pneumoniae.

Clinical indicators in calves experimentally infected with different concentrations of Streptococcus pneumoniae (n = 9). Table 2

Groups	Examination time, days	Body temperature, ° C	Heart rate (beats per minute)	Respiratory rate (breaths per minute)
Group I (Experimental)	At the beginning of the experiment	39,5±0,2	119,0±4,0	28,0±0,5
	2	39,9±0,4	126,0±4,0	30,0±0,5
	5	41,9±0,3	132,6±3,4	32,6±2,2****
	7	41,0±0,2	136,0±4,0	38,0±0,5
M±m		40,9±0,24	131,3±4,74	33,3±0,42
Group II (Experimental)	At the beginning of the experiment	38,6±0,6	120,0±4,4	28,0±0,3

	2	41,1±0,4**	128,0±4,2	33,2±0,4
	5	40,9±0,5	134,6±3,2	38,1±2,8
	7	41,2±0,3	132,0±4,4***	37,0±0,3
M±m		41,07±0,8	131,3±3,36	36±0,43
Group III (Control)	At the beginning of the experiment	38,4±0,3	115,0±3,2	29,0±0,4
	2	38,8±0,3	117,0±3,4	29,0±0,4
	5	38,2±0,8	116±2,8	27,0±0,6
	7	38,0±0,3	114,0±3,4	28,1±0,4
M±m		38,3±0,7	115,7±2,11	28,03±0,5

indicates p < 0.05; ** indicates p < 0.001.

According to the results presented in Table 2, in 7–30-day-old calves infected with *Streptococcus pneumoniae* at a dose of 2 × 10^9 microbial cells, clinical parameters were monitored on days 2, 5, and 7 of the experiment. It was determined that, compared to baseline values, body temperature increased on average by 1.4 ± 0.0062°C, respiratory rate increased by 5.3 ± 0.031 breaths per minute, and heart rate increased by 12.3 ± 0.35 beats per minute.

In the second experimental group (1–3-month-old calves) infected with *Streptococcus pneumoniae* at a dose of 3 × 10^9 microbial cells, the following clinical changes were observed: although body temperature was within the normal range at the beginning of the experiment, it increased by an average of 2.47 ± 0.03°C during the study. The heart rate increased by 11.3 ± 0.3 beats per minute, and the respiratory rate increased by 8 ± 0.11 breaths per minute compared to baseline values.

In contrast, clinical indicators in calves of different ages in the control group remained unchanged.

During the experiments, blood samples were collected from calves infected with *Streptococcus pneumoniae* at the beginning of the study, as well as on days 3 and 5 of the experiment. The samples were analyzed using the leukocyte differential count to determine morphological parameters, and the results were compared with baseline blood values.

Compared with baseline values, leukocytes, band neutrophils, and segmented neutrophils increased, whereas eosinophils, basophils, lymphocytes, and monocytes decreased. These findings indicate that acute pathogenic *Streptococcus pneumoniae* infection leads to altered reactivity in the organism of young calves.

The housing conditions of the experimental calves, clinical signs, body temperature, heart rate per minute, respiratory rate per minute, and morphobiochemical blood parameters were systematically monitored. During the course of the experiment, blood samples were collected from the calves at the beginning of the study and on days 3 and 5, and hematological as well as biochemical parameters were analyzed at BIOQON Laboratory.

The obtained results were compared with baseline values. The following biochemical parameters were determined: total protein, albumin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), amylase, lactate dehydrogenase (LDH), bilirubin, creatinine, calcium, triglycerides, gamma-glutamyl transferase (GGT), glucose, lipids (fat), magnesium, phosphorus, urea, and other biochemical indicators (Table 4).

Changes in biochemical blood parameters in calves of different ages experimentally infected with Streptococcus pneumoniae over a 5-day period (n = 9) **(Tabel 3)**

Indicators	**Group I (Experimental)** – 7–30 days old	**Group II (Experimental)** – 1–3 months old	**Group III (Control)** – 30, 60, and 90 days old

Pre-experimental values			
Albumin, g/L	27,03±0,5	26,22±1,35	29,10±0,45
Total protein, g/L	65,74±3,10*	62,84±2,51	55,34±1,69
Urea, mmol/L	4,89±0,14	5,56±0,29	3,94±0,15
ALT (alanine aminotransferase), U/L	59,14±3,15	61,93±1,82	76,58±3,49
Amylase, U/L	89,56±4,12	80,10±3,45	78,74±4,21
Creatinine, µmol/L	77,16±3,43	85,64±4,25	89,39±4,52
ALT, U/L *(duplicate—keep one in your table)*	25,33±1,11	23,12±0,45	26,03±1,25
AST (aspartate aminotransferase), U/L	65,34±1,25	68,12±3,19	60,45±2,14
Total bilirubin, mmol/L	5,1±0,2	5,6±0,28	4,8±0,19
Calcium, mmol/L	2,8±0,09	2,1±0,04	2,5±0,2
Cholesterol, mmol/L	2,70±0,05	2,76±0,1	2,80±0,2
GGT (gamma-glutamyl transferase), U/L	9,8±0,6	9,8±0,27	9,9±0,25
Glucose, mmol/L	2,5±0,08	4,0±0,11	3,7±0,05
Lipids (fat), µmol/L	28,6±0,6	20,2±0,8	25,8±0,3
LDH (lactate dehydrogenase), U/L	930,9±14,2	924,4±16,1	932,5±15,7
Magnesium, mmol/L	1,2±0,07	1,0±0,04	0,9±0,09
Triglycerides, mmol/L	0,82±0,03	0,78±0,02	0,80±0,04

Alkaline phosphatase, U/L	34,8±2,10	35,2±1,28	36,3±2,35
Biochemical alterations on day 3 following experimental infection with Streptococcus pneumoniae.			
Albumin, g/L	18,4±1,02	18,33±2,10	29,03±2,12
Total protein, g/L	72,3±3,58	71,27±3,09	55,34±3,69
Urea, mmol/L	5,02±0,22	2,8±0,08	3,94±0,25
ALT (alanine aminotransferase), U	62,16±2,41	22,67±2,10	76,58±3,49
Amylase, U/L	85,33±5,14	72,62±4,11	67,46±3,56
Creatinine, µmol/L	172,3±5,00	164±7,33	89,39±4,58
ALT, U/L	24,3±0,34	22,67±0,78	26,21±1,27
AST (aspartate aminotransferase), U/L	122,0±4,7	143±5,0	60,90±4,0
Total bilirubin, mmol/L	3,73±0,04	3,43±0,03	4,82±0,11
Calcium, mmol/L	1,7±0,02	2,5±0,01	2,61±0,16
Cholesterol, mmol/L	2,48±0,09	3,54±0,14	2,80±0,12
GGT (gamma-glutamyl transferase), U/L	11,2±1,04	11,4±0,71	9,9±0,25
Glucose, mmol/L	2,1±0,02	2,2±0,01	3,7±0,05
Lipids (fat), µmol/L	35,57±1,21	37,9±1,85	28,8±1,15
LDH (lactate dehydrogenase), U/L	1529,0±65,78	1464±71,5	932±10,4
Magnesium, mmol/L	3,66±0,24	3,58±0,20	0,9±0,07

Triglycerides, mmol/L	1,07±0,03	1,02±0,04	0,80±0,02
Alkaline phosphatase, U/L	45,37±3,38	76,80±5,39	36,34±2,54
Biochemical alterations on day 5 following experimental infection with Streptococcus pneumoniae.			
Albumin, g/L	14,7±0,32	19,21±1,04	29,03±2,12
Total protein, g/L	84,3±3,04	82,5±3,52	55,34±3,69
Urea, mmol/L	6,04±0,38	6,18±0,28	3,94±0,25
ALT (alanine aminotransferase), U	68,21±2,6	67,22±3,15	76,58±3,49
Amylase, U/L	81,7±2,58	70,15±3,45	67,46±3,56
Creatinine, µmol/L	181,14±4,66	178,4±5,02	89,39±4,58
ALT, U/L	25,6±2,7	25,8±1,09	26,21±1,27
AST (aspartate aminotransferase), U/L	128,8±3,4	151,3±4,68	60,90±4,0
Total bilirubin, mmol/L	4,28±0,12	7,26±0,5	4,82±0,11
Calcium, mmol/L	1,4±0,01	1,3±0,11	2,61±0,16
Cholesterol, mmol/L	2,74±0,15	3,92±0.21	2,80±0,12
GGT (gamma-glutamyl transferase), U/L	12,4±1,12	12,8±1,10	9,9±0,25
Glucose, mmol/L	2,0±	1,9±0,01	3,7±0,05
Lipids (fat), µmol/L	36,94±2,3	38,88±2,28	28,8±1,15
LDH (lactate dehydrogenase), U/L	1624,8±43,2	1557,6±38,2	932±10,4

Magnesium, mmol/L	4,11±0,13	4,10±0,2	0,9±0,03
Triglycerides, mmol/L	1,01±0,04	1,04±0,06	0,80±0,02
Alkaline phosphatase, U/L	55,22±2,46	79,45±4,18	36,34±2,54

indicates p < 0.05; ** indicates p < 0.001.

Biochemical analyses of blood samples obtained from calves infected with Streptococcus pneumoniae were performed using the methods of Gornall, Doumas, Reitman–Frankel, Trinder, Jaffe and others. These analyses were conducted in the biochemistry and serology laboratory of BIOQON Laboratory.

According to the results presented in Table 4, in the first experimental group (calves infected with 2 × 10^9 microbial cells), the following changes were observed on day 3 of the experiment: albumin decreased by 8.63 ± 0.12 g/L, total bilirubin by 1.37 ± 0.12 mmol/L, and calcium by 1.1 ± 0.02 mmol/L compared to normal values. At the same time, other biochemical parameters increased, including total protein (6.56 ± 0.21 g/L), urea (0.13 ± 0.004 mmol/L), creatinine (95.14 ± 3.82 µmol/L), aspartate aminotransferase (AST) (56.66 ± 0.58 U/L), gamma-glutamyl transferase (GGT) (1.4 ± 0.03 U/L), and lactate dehydrogenase (LDH) (598.1 ± 14.26 U/L).

These changes indicate that the acute pathogenic effect of Streptococcus pneumoniae infection in 7–30-day-old calves led to dehydration, which in turn caused increases in urea and creatinine levels.

In the second experimental group (calves infected with 3 × 10^9 microbial cells), biochemical analysis on day 3 revealed that albumin decreased by 2.11 ± 0.15 g/L, total bilirubin by 2.17 ± 0.12 mmol/L, and calcium by 0.4 ± 0.01 mmol/L. Meanwhile, total protein increased to 8.43 g/L, urea to 2.76 ± 0.12 mmol/L, creatinine to 78.36 ± 3.04 µmol/L, AST to 74.88 ± 4.24 U/L, GGT to 1.6 ± 0.02 U/L, and LDH to 539.6 ± 6.87 U/L.

On day 5 of the experiment, in the first experimental group (2 × 10⁹ dose), albumin decreased by 12.33 ± 1.09 g/L, total bilirubin by 0.82 ± 0.08 mmol/L, and calcium by 1.4 ± 0.02 mmol/L. At the same time, increases were observed in total protein (18.56 ± 0.39 g/L), urea (1.15 ± 0.02 mmol/L), creatinine (89.14 ± 1.91 mmol/L), AST (63.46 ± 2.04 U/L), GGT (2.6 ± 0.12 U/L), and LDH (693.9 ± 8.31 U/L).

In the second experimental group (1–3-month-old calves infected with 3 × 10⁹ colony-forming units), biochemical analysis showed that serum albumin decreased to 7.01 ± 0.18 g/L, while alkaline phosphatase activity remained nearly unchanged in all groups. In contrast, total protein increased to 19.66 ± 0.18 g/L, and similar increases were observed in urea, creatinine, and alanine aminotransferase (ALT), likely due to stress-related effects in the infected calves.

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