

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Clinical And Biological Aspects Of Horse Pyroplasmidosis In The South Of Russia.

Svetlana Nikolaevna Lutsuk*, Maria Evgen'evna Ponomareva, Yuliya Vasil'yevna Dyachenko, Vladimir Aleksandrovich Orobets, and Bagama Manapovich Bagamaev.

Stavropol State Agrarian University, Zootekhnicheskiiy lane 12, Stavropol 355017, Russia

ABSTRACT

The article presents the results of observation of clinical signs in foals with pyroplasmidosis, which showed a different course of the disease. Increasingly, horse teileriosis occurs chronically and is accompanied by edema and bronchopneumonia. Studies show that in the body of patients with pyroplasmidosis of animals, metabolic changes are directed toward reducing the activity of oxidation-reduction processes, as a result of which biosynthesis is slowed down and enzyme activity is increased. The use of food additive from the larvae of drones has a beneficial effect on the organism of horse-carriers.

Keywords: Babesiaballi, Nuttalliaequi, Theileriaequi, biochemical indicators of horses, hematological indicators of horses, biologically active additive, larvae of drone, by-products of beekeeping.

**Corresponding author*

INTRODUCTION

Pyropamidosis of horses is a widespread vector-borne disease caused by protozoa *Piroplasmida: Babesiacaballi* and *Nuttallia (Theileria) equ* [10, 11]. Pyroplasmidosis - systemic diseases. Typically, the appearance of signs of heart failure, stagnation [2], anemia, jaundice, erythrocyte hyperchromia, dyspnea, gastrointestinal disturbances [11, 12]. A decrease in the total protein, a sharp decrease in β -globulins and an increase in γ -globulins, an increase in total and direct bilirubin, a reduction in glucose concentration, an increase in the activity of transamination enzymes [3,4], thrombocytopenia, a decrease in the number of erythrocytes, hematocrit and hemoglobin, leukopenia, lymphopenia, neutropenia and monocytosis. [5, 6]. The analysis of the disease course of foals in a permanently unfavorable place is carried out, changes in biochemical and hematological indices of blood of horses bearing pyroplasmidosis and the scheme of supporting and restoring treatment of convalescents.

MATERIALS AND METHODS

Every day inspections of the cows and their foals were conducted, the number of ticks recorded, their appearance determined. Have observed the course of the disease. Every day we examined the peripheral blood smears, colored according to Romanovsky-Giemsa, 200 fields of vision per smear with an increase of 70-90. The intensity of parasitemia was expressed as a percentage of the total number of erythrocytes.

Blood of sick animals was examined by hematological parameters, serum for total protein and protein fractions, glucose, activity of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase.

The determination of bilirubin in the serum was performed using the Jendrassik method with the caffeine reagent. Transaminases and urea were determined by the kinetic method on the biochemical analyzer Airone-200-RA, creatinine by the kinetic method according to Jaffe, glucose by the glucose oxidase method. Hematologic studies were performed on the hematological analyzer Systex-kx-21. The total protein content was determined by the refractometric method, the protein fractions by the nephelometric method.

To determine the effect of the biologically active additive on the organism of convalescent animals, 10 carrier horses were divided into two groups of 5 heads: the first was fed from the homogenized larvae of honey bee drone and carrots for 7 days. The second group was given just carrots in the same amount. Biologically active additive from larvae of drones was prepared according to the prescription given in the patent [4]. Hematologic and biochemical parameters were determined before the feeding of the additive and after the 7-day course was completed.

Data processing was performed using the MicrosoftExcel statistical analysis package using the Student's test.

RESULTS AND DISCUSSION

The territory of the stud farm, where we conducted research, is characterized by a strong annual fluctuation in the number of ticks and diseased animals. According to our observations, in different years during the seasonal outbreaks (in May-June) there were 36.6 to 75.3% of the youngsters of the current year of birth. The disease in them proceeded in different ways (Table 1).

The sick colts of group I, when the herd returned from the pasture, walked behind everyone with their head down, in the left they were half-closed, legs wide apart. Visible mucous membranes were initially hyperemic, and then, on the second day, pale with icteric tinge. The heart beat became pounding, the pulse uneven, breathing difficult. Within 3-4 days the temperature and parasitemia remained within the same limits. In the smears of peripheral blood, small and rare nuttalliums were found in the form of "Maltese crosses", commas, rounded and annular forms. No other signs of the disease were observed.

Table 1: Temperature and parasitic response of horses spontaneously ill with nuttalliosis

Group	Index		Before the disease	Days of illness								
				1	2	3	4	5	6	7	8	9
I group	body temp., °C	M	38,3	39,3	39,5	39,3	39,5	–	–	–	–	–
		m	0,23	0,88	0,19	0,88	0,00	–	–	–	–	–
	parasitemia, %	M	0,00	6,82	7,30	7,33	8,05	–	–	–	–	–
		m	0,00	0,33	0,19	0,33	0,66	–	–	–	–	–
II group	body temp., °C	M	38,2	39,2	38,7	38,5	38,5	38,5	38,5	38,7	38,7	38,4
		m	0,35	0,57	0,16	0,16	0,01	0,01	0,01	0,23	0,16	0,01
	parasitemia, %	M	0,00	3,76	0,33	0,1	0,1	0,10	0,10	1,20	3,86	0,10
		m	0,00	0,33	0,23	0,01	0,01	0,01	0,00	0,90	0,57	0,01
III group	body temp., °C	M	38,3	39,5	39,0	39,1	39,8	38,9	38,5	38,5	–	–
		m	0,15	0,89	0,89	0,10	0,90	0,14	0,24	0,01	–	–
	parasitemia, %	M	0,00	7,80	7,60	8,18	8,62	6,50	0,64	0,10	–	–
		m	0,00	0,84	0,22	0,32	0,17	1,50	0,26	0,01	–	–

The foals of group II on the first day of the disease showed a slight increase in temperature - up to 39.2±0.57 ° C, parasitemia was 3.76-0.33%, slight depression and appetite worsened. However, on the second day, the temperature dropped to normal, and in peripheral blood smears only single parasites were detected, after 6-7 days the temperature rose again to 38.7±0.2 °C, the number of parasites increased to 3.86±0.57% , the general condition worsened slightly. This condition was observed in foals for two days, and then they recovered.

In the foals of group III, clinical signs similar to those in group I were observed in the first two days. On the third day, the foals started coughing, at first auscultation in the lungs was heard dry, and on the 4th-5th day - wet rales. Body temperature remained elevated (39.82±0.9 °C), persistent fever of a constant type developed within 5-6 days. The condition of foals every day worsened, all the clinical signs of bronchopneumonia developed, which were treated symptomatically.

Some biochemical indices of the blood of sick horses were studied (Table 2).

Table 2: Some biochemical parameters of blood of horses with pyroplasmosis

Group		Healthy M±m	Sick M±m
AST, µkat / l		0,05±0,004	0,25±0,031
ALT, µkat / l		0,051±0,014	0,10±0,007
Alkaline phosphatase, µkat / l		1,11±0,019	1,19±0,016
Glucose, mmol / l		5,45±0,093	4,85±0,011
Total protein, g / l		63,77±0,526	55,83±0,335
Albumins, g / l		34,91±0,249	30,85±0,460
Globulins, g / l	α	11,46±0,124	12,23±0,225
	β	11,11±0,079	11,73±0,194
	γ	5,27±0,021	4,2±0,013
Urea, mmol / l		–	3,48±0,08
Residual nitrogen, mmol / l		–	1,47±0,06

In patients with foals, the amount of total protein is reduced by 17%, glucose by 12% (p<0.001), which may indicate a higher level of energy consumption in sick animals. The activity of transamination enzymes (ALT and AST) in patients with animals is much higher (p<0,001) than in healthy ones (2 and 5 times, respectively), which is observed in liver lesions (ALT) and heart (AST), as well as necrotic processes in the body.

Carrying parasites, often after recovery, is not indifferent to animals. In horses it is manifested not by pronounced changes in blood parameters: thrombocytopenia, neutropenia, monocytosis, increased activity of AsAT [5].

In order to improve the condition of the animals carrying pyroplasmids, we used a biologically active additive from the larvae of drones (Table 3).

Table 3: Hematologic parameters of horses of patients with pyroplasmidosis before and after application of dietary supplements

Index	Blood test results				N
	before applying biologically active additive		after applying biologically active additive		
	1 group	2 group	1 group	2 group	
Leucocytes, 10 ⁹ / l	7,8±1,15	7,4±0,26	9,7±1,34	7,3±0,16	7-11
Erythrocytes, 10 ¹² / l	7,8±0,37	7,6±0,30	7,9±0,42	7,5±0,28	6-9
Hemoglobin, g / l	129,4±10,24	127,4±4,51	122,0±7,97	125,4±5,53	80-140
Hematocrit, %	38,0±3,00	39,0±1,00	37±2,00	38,0±1,00	30-50
Average volume of erythrocyte, mkm ³	47,9±1,70	51,5±0,75	46,6±1,42	48,5±0,75	34-58
Average concentration of Hb in erythrocyte, %	16,4±0,62	18,0±0,36	15,4±0,57	17,0±0,41	31-37
Average content of Hb in erythrocytes, pg	343,8±6,76	350,0±2,61	331,0±3,18	348,0±2,34	10-18
Platelets, 10 ⁹ / l	127,8±4,84	118,2±17,19	158,8±10,94	119,2±10,10	200-500
ESR, mm/h	32,0±12,23	35,4±6,05	27,6±9,64	33,4±3,05	up to 60
Eosinophils, %	1,6±1,36	1,0±0,45	3,4±0,87	1,4±0,55	0-4
Stool neutrophils, %	1,4±0,51	2,2±0,2	2,8±0,97	2,0±0,3	0-6
Segmented neutrophils, %	45,2±4,68	47,6±2,58	52,0±2,39	46,7±2,73	54-65
Lymphocytes, %	45,4±4,25	44,0±3,45	38,6±1,78	44,6±3,05	16-43
Monocytes, %	6,6±1,21	6,8±0,49	3,8±0,97	6,7±0,15	1-6

Thrombocytopenia is a characteristic feature of pyroplasmidosis [4, 5]. Platelets accelerate the healing of wounds and restore damaged internal organs, increase the phagocytic function of leukocytes, including natural killers [1]. Thus, an increase in the number of platelets indicates an increase in the general resistance of the body and its reparative functions. Normalization of the number of neutrophils and monocytes may indicate an increase in the protective functions of the body. These indicators tend to normalize in horses after using an additive from the larvae of drones. The number of platelets in the first group significantly increased by 24.3% (p < 0.05).

The results of biochemical studies of horse disease before and after the application of the biologically active additive are given in Table 4.

Table 4: Biochemical blood indices of horse-carriers of pyroplasmids before and after application of biologically active additive

Index	Blood test results				N
	before applying biologically active additive		after applying biologically active additive		
	1 group	2 group	1 group	2 group	
Bilirubin total, μmol / l	34,8±12,24	30,7±2,38	34,5±10,06	31,7±3,28	5,4-51,4
AsAT, U / l	273,6±18,54	260,8±11,67	305,3±14,01	263,4±15,67	130-300
AlAT, unit / liter	18,1±1,20	14,0±0,93	17,0±0,79	15,0±1,84	2,7-20
Glucose, mmol / l	4,6±1,05	5,1±0,22	5,3±0,16	5,0±0,17	3-7
Urea, mmol / l	5,8±1,02	5,0±0,32	6,4±0,54	5,2±0,21	3,5-8,8
Creatinine, μmol / l	132,6±11,76	148,1±10,77	104,7±5,86	147,1±8,75	80-180

Index	Blood test results				N
	before applying biologically active additive		after applying biologically active additive		
	1 group	2 group	1 group	2 group	
Total protein, g / l	62,4±3,06	66,4±1,32	63,9±1,45	66,1±1,42	47-75
Amylase, U / l	11,8±5,15	6,9±2,3	15,5±3,66	7,8±1,3	4,9-16,5
Timole sample, unit.	2,5±0,63	2,1±0,69	2,9±0,56	2,1±0,75	1-5

Attention is drawn to the significant and reliable (by 26.6%, $p < 0.05$) decrease in the level of creatinine after giving horses an additive from the larvae of drones. Pyroplasmidosis in horses is accompanied by a lesion of the excretory system, sometimes extremely acute [8]. Lowering the level of creatinine indicates the normalization of kidney activity.

Thus, the use of a feed additive from the larvae of drones has a beneficial effect on the body of horse-carriers of pyroplasmids.

CONCLUSION

Thus, depending on various factors, we note the adaptation of the pathogen, which manifests itself in a change in the apparent sizes of pathogens detected by microscopy of smears, as well as in the change in their virulence, which manifests itself in a different course of the disease. In recent years, the theileriosis of horses is increasingly chronic and is accompanied by edema and bronchopneumonia. We believe that further studies of this process are required, including a study of the genetic structure of the causative agent population in the North Caucasus region.

In addition to specific treatment, the use of fodder additive from the larvae of drones has a beneficial effect on the body of horse-carriers and leads to a shift in blood levels from extreme to average values of the norm.

REFERENCES

- [1] Danilov, I.P. Platelets: a new look at their role in the body // *Meditsinskiye novosti*. 2008. №9. p. 17-19.
- [2] Lutsuk, S.N., Ponomareva, M.E. *Pyroplasmidosis of horses: monograph – Edition 2, revised* - Stavropol: Publishing house "AGRUS", 2004. - 152 p.
- [3] Ponomareva, M.E., Lutsuk, S.N. Some biochemical indicators of blood of horses with pyroplasmidosis // *Materials of IV International conference «Aktual'nyye problemy veterinarnoy meditsiny melkikh domashnikh zhivotnykh i loshadey»* (May 31, 2001). - Stavropol, 2001. - P. 99-101.
- [4] Lutsuk S.N., Zhukova N.S., Ponomareva M.E., Khodusov A.A. Method of preparation of a biologically active additive for animals and a product on its basis. // Patent for invention RUS 2533156 23.07.2013.
- [5] Lutsuk S.N., Zhukova N.S., Ponomareva M.E. Biochemical and hematological status of horses - carriers of babesias and the influence of biologically active additive from larvae of drones on it // *Vestnik APK Stavropol'ya*. 2013. № 2 (10). p. 216-218.
- [6] Adam M., Pikalo J., Snyder A., Steinrigl A., Köller G., Schusser G.F. Equine Piroplasmosis – a case of severe Babesiababalis infection associated acute renal failure // *Berliner und MünchenerTierärztlicheWochenschrift*. 2017. V. 130, №3/4, p. 113–118.
- [7] Fabisiak, M., Sapierzynski, R., Klucinski, W. Analysis of haematological abnormalities observed in dogs infected by a large Babesia // *Bulletin of the veterinary institute in Pulawy*. – 2010. – V. 54, № 2. – p. 167-170.
- [8] Kirtz, G., Leschnik, M., Hooijberg, E. In-clinic laboratory diagnosis (Babesiacaniscanis) for veterinary practitioners in Central Europe // *Tieraerztliche praxis ausgabe kleintiereheimtiere*. – 2012. – V. 40, №2. – p. 87-94.
- [9] Lutsuk S.N., Ponomareva M.E., Tolokonnikov V.P., DyachenkoYu.V., TokhovYu.M., Orobets V.A. Enzootic peculiarities of horse's babesiosis in the North-Caucasus region // *Research Journal of Pharmaceutical, Biological and Chemical Sciences*. 2016. T. 7. № 6. p. 2295-2298.



- [10] Waal D. Equine piroplasmiasis: a review // Br Vet J. 1992. № 148. p. 6–14.
- [11] Wise L., Kappmeyer L., Mealey R., Knowles D. Review of equine piroplasmiasis // J Vet Intern Med. 2013. № 27. p. 1334–1346.
- [12] Zobba R., Ardu M., Niccolini S., Chessa B., Manna L., Cocco R., Parpaglia M. Clinical and laboratory findings in equine piroplasmiasis // J Equine Vet Sci. 2008. № 28. p. 301–308.