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Comparative Characteristics of Treatment Methods in Dogs Isosporosis and Giardiasis

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ABSTRACT

Gastrointestinal tract diseases in dogs retain a leading position among other pathologies. At the same time, gastrointestinal parasites are among the most common pathogenic agents in dogs around the world and make a significant contribution to the development of severe gastroenteritis. Giardia sp. is a common cause of acute gastroenteritis in many animal species around the world. Proven participation of Giardia sp. in the development of chronic gastrointestinal disorders, leading to malabsorption and developmental delay in young animals, and, as a result, a decrease in resistance and an increase in susceptibility to reinvasion by giardia and various infections. In this regard, it seems relevant to study the pathogenetic features of the effect of protozoa on the mucous membrane of the small intestine of dogs for the development and improvement of therapeutic regimens. In our work, the therapy of giardiasis in domestic dogs with Drontal plus reinvasion for 6 months was not observed. When treating dogs with giardiasis and cystoisosporosis, the main antiprotozoal drug (Drontal for giardiasis or Stop-Coccid for cystoisosporosis) + antibiotic Cobactan + Probiotic Pro-Colin + vitamin B complex (Milgamma), the dogs were restored 100% on the 30th day after the end of treatment. During long-term follow-up of patients with clinical manifestations of gastrointestinal tract malfunction (a change in the consistency of feces towards softening, refusal to feed, and vomiting) with the use of a prebiotic was not observed.

Keywords: Biochemical Blood Test, Complete Blood Count, Cystoisosporosis, Dogs, Giardiasis, Invasion.

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INTRODUCTION

With the occurrence of protozoal invasions in the animal organism, an important role is played by processes provoked by secondary factors or being secondary. In this regard, the invasion process cannot be considered only as a local phenomenon. It is important to approach as a disease of the whole organism as a whole. The blood composition in the body and the gastrointestinal tract are sensitive systems that show the effect and its degree on the body of various factors [1, 2, 3]. Digestion and absorption of nutrients in the gastrointestinal tract requires a complex interaction between motor, secretory, digestive and absorbing functions. The proximal small intestine absorbs nutrients extremely efficiently: after perfusion of nutrients into the duodenum, up to 80% of triglycerides, 60% of carbohydrates and 50% of proteins are absorbed during physiological processes and the loss of functioning of the small intestinal mucosa can impair their absorption [4, 5].

The mucous membrane of the small intestine is a tissue with a high level of cellular renewal. With toxic effects, the balance between proliferation and cell death is disturbed, which leads to morphofunctional changes in the tissues. In the mucous membrane of the small intestine, a violation of the intestinal absorption of nutrients, malabsorption syndrome occurs. Infectious and parasitic factors are often the etiological factor in the development of secondary or acquired malabsorption [6, 7].

The main reasons for its development are more often associated with insufficiency of intestinal and pancreatic enzymes, substrate-binding proteins, intestinal and very parietal digestion disorders, as a result of which the motility of the entire gastrointestinal tract is impaired, intestinal villus atrophy occurs. As a rule, all these changes can cause protozoa with parasitizing in the intestine [8].

Protozoa often cause the development of pathologies of the gastrointestinal tract in dogs. Most protozoal pathogens are associated with cystoisospores and giardia.

Giardia sp. often cause the development of disorders of the gastrointestinal tract, especially absorption processes in the small intestine. In the clinical picture of giardiasis, malabsorption syndrome plays a leading role, however, the issues of correcting impaired intestinal absorption are not adequately covered in the scientific literature. In some cases, malabsorption can be implicit, manifest as a pathology of other organs of the systems: digestion (liver, pancreas), respiration (lungs) and hematopoietic (anemia), as well as systemic problems - growth retardation. Due to the violation of the intake of macro- and micronutrients, "deficient" conditions develop, the clinical manifestations of which create difficulties for the timely assessment of the

secondary pathological process, worsen the prognosis and contribute to a long recovery period [9, 10, 11].

Clinically significant invasion mainly occurs in young animals, but the clinical signs of pathogens vary from asymptomatic to severe enterocolitis [12, 13].

Malabsorption syndrome occurs as a result of damage to the structure of the mucous membrane of the small intestine under various pathological conditions of the gastrointestinal tract and other organs and systems (enteritis, diabetes mellitus, cholestasis, impaired arteriomesenteric circulation, toxic effects on the mucous membrane of the small intestine, impaired intestinal motility, HIV infection, intestinal dysbiosis, chronic pancreatitis, etc.) [14, 15].

It has been established that the clinical manifestations of malabsorption are almost identical for all diseases of the small intestine and are expressed in metabolic disorders: protein, water-electrolyte, hypovitaminosis, iron deficiency anemia. The pathogenetic mechanisms of the development of strategic offensive arms significantly differ with individual nosological forms. With celiac disease, the main cause of malabsorption is a decrease in the absorption surface of the small intestine due to mucosal atrophy. Among the reasons for the development of a syndrome of impaired absorption: bacterial seeding of the small intestine, giardiasis, which develops as a result of impaired secretory IgA production in the small intestine mucosa. As a result, damage to the mucous membrane and a violation of membrane digestion occur [16].

The aim of the study was to evaluate the effectiveness of various treatment regimens for hyardiosis and cystoisosporosis in dogs.

MATERIALS AND METHODS

The work was carried out in 2014-2020 in the Department of Veterinary Medicine of the Agrarian and Technological Institute of the Peoples' Friendship University of Russia (RUDN University) and the Department of Parasitology and Veterinary and Sanitary Expertise at the Moscow State Academy of Veterinary Medicine and Biotechnology - MVA named after K.I. Scriabin.

The clinical part was performed in Moscow clinics. In total, 343 domestic dogs of different breeds and ages were examined. For endoscopic examination, 22 dogs were kept in a shelter for homeless animals in the Moscow Region: 7 animals infected with Cystoisospora sp., 15 dogs infected with Giardia sp.

Research and registration of clinical status was carried out according to generally accepted methods [17]. Blood for the study was taken from animals before feeding after 8 hours of a hungry diet, in the morning from vena saphena into 3 plastic tubes. For hematological studies with a volume of 2 ml, for biochemical studies - with a volume of 3 ml.

Dogs with clinical manifestations of gastrointestinal tract malfunction were grouped according to the principle of analogues.

Blood samples were taken from 53 dogs under the age of 3 years, including 19 dogs with Giardia sp., 27 with Cystoisospora sp., 7 dogs with mixed invasion of Giardia sp. and Cystoisospora sp. The control group included 8 clinically healthy dogs aged 1.5-2 years with a negative parasitological study.

A complete blood count was performed on a ABC VET hematology analyzer (France). The morphological features of red blood cells were determined by the change in their diameter, thickness and shape. The leukogram was counted in blood smears stained according to Romanovsky – Giemsa [18]. Data was compared with Harvey J.W. Diagnostic Guide and Color Atlas. (2012) [19].

Blood serum was obtained by whole blood sedimentation and blood clot retraction followed by centrifugation on a Liston C2204 centrifuge (1.0; 1.5; 2.0; 3.0 thousand rpm 12x15 ml) (Biosan, Latvia) at 2000 rpm for 10-15 minutes [20]. Blood serum was examined for 4 hours. Hemolized and chylous samples were not used. The biochemical composition of blood serum was studied using a Humalizer Junior automatic biochemical analyzer (HUMAN, Germany). The results were evaluated in accordance with the recommendations of the management of Barger A.M., MacNeill A.L. (2015) [20].

Blood was taken from all animals at diagnosis and on the 30th day after the end of therapy.

The first experiment was to compare the effectiveness of the use of an antibacterial drug or probiotic as a concomitant drug in the treatment of cystoisosporosis and giardiasis.

Dogs with giardiasis were divided into 3 groups:

Group 1 - (9 dogs) received Drontal® Plus (Waueg Animal Health GmbH, Germany) for three days at a dose of 1 tablet per 10 kg of animal body weight and the antibacterial drug Cobactan 2.5% was used as additional therapy (MSD Animal Health, USA) intramuscularly at a dose of 1 ml per 10 kg of animal body weight 1 time per day for 6 days (according to the manufacturer's instructions).

Group 2 - (9 dogs) received Drontal® plus according to the scheme presented above, and a pro-coli feed supplement with Pro-Colin (Probiotics International Ltd., UK) at a dosage of 3-5 ml 2 times a day 30 days on an empty stomach 30-40 minutes before feeding (according to the manufacturer's instructions).

3rd group - (7 dogs) were treated only with Drontal plus according to the scheme presented above.

Dogs invaded by cystoisosporosis were also divided into 3 groups:

Group 4 - 9 dogs received Stop- Coccid (Api-San LLC, Russia) (toltrazuril) at a dose of 10 mg / kg once a day for 3 days orally and intramuscularly - Cobactan 2.5% (MSD Animal Health, USA) at a dose of 1 ml per 10 kg of body weight 1 time per day for 6 days.

5th group - 9 dogs received (Stop-Coccid preparation, LLC NPI Api-San, Russia) (toltrazuril) according to the scheme presented above, and a feed supplement with Pro-Colin prebiotic at a dosage of 3-5 ml 2 times a day 30 days.

6th group - 6 dogs were treated only with toltrazuril [10].

The second series of experiments consisted in comparing two complex treatment regimens: 1. the main antiprotozoal drug + antibiotic + probiotic and 2. the main antiprotozoal drug + antibiotic + probiotic + complex B vitamins (Milgamma, Worwag Pharma GmbH & Co. KG (Germany), solution for intramuscular administration).

To conduct this study, 3 groups of dogs were formed that were spontaneously infected with giardiasis: the 7th group, consisting of 6 dogs, received therapy according to the scheme - Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin; The 8th group (6 dogs) received therapy according to the scheme - Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma; The 9th group (4 dogs) was treated only with the Drontal plus antiprotozoal drug and was a control.

Dogs spontaneously infected with cystoisosporosis were also divided into 3 similar groups: the 10th group (5 goals) received therapy, including the use of Stop-Coccid + Cobactan 2.5% + Pro-Colin prebiotic; The 11th group (5 goals) was treated according to the scheme - Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma; The 12th group (4 heads) was treated only with Stop-Coccid and was a control.

All animals participating in the experiment were transferred to Purina EN therapeutic feed for 6 months. Changes in the clinical condition of animals were assessed by: the presence / absence of diarrhea, a change in the consistency of feces, the presence / absence of mucus and / or blood in the feces, a change in appetite, and the presence of vomiting for 6 months.

The reliability of the results relative to each other and relative to the norm, as well as the statistical significance of intergroup differences, was evaluated using the standard student criterion. Values of P <0.05 were considered significant. Nonparametric methods were chosen to increase the reliability of the comparison in the presence of small samples. The values of the results obtained are presented in the form of the average value and standard error of the mean (M ± m). All analyzes were performed using SPSS software for Windows version 2.0.

RESULTS AND DISCUSSION

The first experiment consisted in comparing efficacy, as concomitant therapy for cystoisosporosis and giardiasis, the use of an antibacterial drug or probiotic. The results of hematological and biochemical studies are shown in tables 1-4.

Table 1: Complete blood count of dogs infested by Giardia sp. during treatment, M±m

Value	Before therapy	30 days after therapy			Reference values
		1st group Drontal® plus + Cobactan 2.5% (n = 9)	2nd group Drontal® plus + Pro-Colin (n = 9)	3rd group Drontal® plus (n = 7)	
RBC, x10^{12}/л	5.7±0.8	6.4±1.4	7.2±2.1	6.0±1.7	5,5-8,5
WBC, x10^{9}/л	22.6±2.3*	11.4±2.5	16.3±2.3	13.6±3.1	6-17
HB, g / l	126.5±3.7	137.1±3.2	145.3±3.4	132.7±3.7	120-180
PLT, x10^{9}/л	343.4±2.4	338.7±3.7	423.6±3.8	421.4±3.9	200-900
HCT, %	39.4±1.7	41.5±1.4	38.4±1.*	39.7±1.5	37-55
ESR, mm/h	4.4±1.4*	2.8±1.4	2.9±1.3	4.1±1.2	2,0-3,5
MCV, fl	62.8±2.2	65.2±2.6	66.4±2.1	63.2±2.9	60-75
MCHC, %	33.6±3.2	33.4±4.2	32.7±3.9	34.6±4.1	32-36
RDW, %	13.9±2.0	14.8±2.2	13.1±2.1	14.4±2.3	11,9-16,0
MCH, pg	22.6±0.5	23.6±0.8	22.4±0.7	24.8±0.6	21-27
Leukogram					
Monocytes, %	6.0±0.1	9.0±0.3	8.0±0.2	6.0±0.1	3-10
Lymphocytes, %	23.0±1.7	22.0±2.6	9.0±1.4	24.0±1.03	12-30
Basophils, %	0	0	1	0	0-1
Eosinophils, %	3.0	3	2	4	2-10
Neutrophils:					
Band neutrophil, %	6.0±0.4*	2.0±0.34	3.0±0.5	3.0±0.5	0-3
Segmented neutrophil, %	62.0±0.9	64.0±2.7	67.0±2.9	63.0±3.0	60-70

Note: Reference values are given by J.W. Harvey (2012)

P < 0,05.

* - differences between the compared values are significant

According to hematological blood parameters in dogs before treatment, the number of leukocytes, ESR and the level of band neutrophils were overestimated. These indicators may indirectly indicate an inflammatory process in the body of dogs with parasitization of protozoa. The number of red blood cells and hemoglobin were also at the lower boundary of the reference values. Against the background of therapy on the 30th day, these changes returned to normal.

In the results of blood chemistry infested by protozoa dogs, the following data were obtained during therapy, presented in table 1.

Before treatment, dogs with giardiasis had a low content of urea, total protein, albumin, globulin and cholesterol, and a

high content of alanine aminotransferase (ALT) and aspartate aminotransferase (AST). After therapy on the 30th day in dogs of the first group with the addition of Cobactan antibiotic therapy, blood counts recovered and reached normal levels, except for albumin, which corresponded to lower reference values (24.2 ± 4.35 g / l). In dogs of the second group, a reduced amount of total protein (53 ± 4.1 g / l), albumin (23.4 ± 4.21 g / l) and cholesterol (2.5 ± 0.67 mmol / l) remained. In the third group, an underestimated value of total protein, albumin, cholesterol and urea was noted (Table 2).

Table 2: Blood biochemical parameters in dogs infested by Giardia sp. during treatment, M ± m

Values	Units	Before therapy	30 days after therapy			Reference values
			1st group Drontal® plus + Cobactan 2.5% (n = 9)	2nd group Drontal® plus + Pro-Colin (n = 9)	3rd group Drontal® plus (n = 7)	
Total bilirubin	μmol / l	3.4±0.64	4.1±0.51	3.2±0.58	4.2±0.31	< 13.5
Direct bilirubin	μmol / l	2.1±0.03	1.2±0.02	1.6±0.02	1.7±0.01	< 5.5
AST	U / L	67.2±5.81*	35.1±3.60*	29.0±7.65	29.0±3.10	over 6 months: 8-42 (up to 6 months: <70)
ALT	U / L	64.0±4.10	31.4±4.32	22.0±5.66	27.0±5.25	10 – 58
Ritis coefficient	units	1.15	1.12	1.30	1.07	1.1 – 1.3
Urea	mmol / l	3.2±0.63	4.14±1.24	4.2±0.34	3.8±0.82	3.5 – 9.2
Creatinine	μmol / l	124.0±14.45	93.17±15.24	117.0±13.70	112.0±14.25	54-138 (44-90 dogs up to 10 kg)
Total protein	g / l	44.0±3.82*	58.4±5.18	53.0±4.14	51.0±3.16	over 6 months: 55-73 (up to 6 months: 44-56)
Albumin	g / l	20.0±2.54	24.2±4.35	23.4±4.21	22.1±3.86	25 – 39
Alkaline phosphatase	U / L	51.0±12.35	48.87±13.36	38.0±12.61	43.0±12.41	over 8 months: 10-70 (up to 8 months: 80-230)
Alpha amylase, total	U / L	871.3± 144.58	1104.5± 256.55	915.0± 231.84	986.0± 211.50	300 -1500 (over 4 months)
Glucose	mmol / l	5.41±0.94	5.65±0.92	5.3±0.93	5.68±0.91	3.3 – 6.3
Cholesterol	mmol / l	2.1±0.67	3.6±0.83	2.5±0.67	2.4±0.66	2.5-6.0
Triglycerides	mmol / l	0.44±0.13	0.77±0.16	0.42±0.13	0.56±0.28	0.15-0.84
LDH	U / L	192.0± 42.31	247.38± 62.0	186.0± 41.68	216.0± 43.85	23 – 220
Globulin	g / l	23.0±3.87	31.0±4.83	27.0±4.81	25±3.20	26 – 44

Note. Reference values are given by A.M. Barger and A.L. MacNeill (2015). P <0.05.
* - differences between the compared values are significant.

Therapy for dogs with invasion Cystoisospora sp. was carried out according to the scheme: 4th group - dogs received the drug Stop-Coccid and Cobactan 2.5% (MSD Animal Health, USA). 5th group - dogs received the drug Stop-Coccid and Pro-Colin. 6th group - 6 dogs were treated only with Stop-Coccid.

Against the background of the therapy of cystoisosporosis on the 30th day, all hematological parameters came into line with the reference values (Table 3).

Table 3: Complete blood count of dogs infested by Cystoisospora sp. during treatment, M ± m

Values	Cystoisospora sp. invasion	30 days after therapy			Reference values
		4th group Stop Coccid + Cobactan 2.5% (n=9)	5th group Stop Coccid + Pro-Colin (n=9)	6th group Stop Coccid (n=6)	
RBC, $x10^{12}$/л	6.9±1.1	6.9±1.6	6.6±1.4	7.1±1.8	5.5-8.5
WBC, $x10^{9}$/л	26.2±2.9*	15.3±1.9	13.5±2.0	12.6±2.2	6-17
HB, g / l	153.2±3.5	138.2±3.2	123.3±3.6	144.9±3.5	120-180
PLT, $x10^{9}$/л	438.7±3.4	389.4±4.4	360.5±3.5	421.1±4.0	200-900

HCT, %	42.9±1.2	41.8±3.3	44.2±3.6	44.5±3.7	37-55
ESR, mm/h	3.1±1.2	3.4±2.0	2.8±1.7	3.0±1.8	2.0-3.5
MCV, fl	64.0±2.7	71.2±3.9	68.7±3.4	66.0±3.1	60-75
MCHC, %	32.2±3.8	31.6±4.0	32.9±3.9	32.6±3.6	32-36
RDW, %	15.1±2.1	12.6±3.2	13.7±2.4	13.7±2.4	11.9-16.0
MCH, pg	24.0±0.7	24.1±3.0	25.3±0.7	24.8±0.6	21-27
Leukogram					
Monocytes, %	8.0±0.3	7.0±0.2	8.0±0.3	4.0±0.1	3-10
Lymphocytes, %	26.0±1.9	21.0±2.1	19.0±1.9	18.0±1.7	12-30
Basophils, %	1	1	0	0	0-1
Eosinophils, %	5*	3	4	2	2-10
Neutrophils:					
Band neutrophil, %	4.0±0.36	2.0±0.04	1.0±0.01	2.0±0.04	0-3
Segmented neutrophil, %	66.0±1.6	66.0±4.2	68.0±4.3	64.0±4.1	60-70

Note. Reference values are given by J.W. Harvey (2012)
P < 0,05.
* - differences between the compared values are significant

Dogs with confirmed cystoisosporosis had lower levels of urea, total protein, albumin, globulin, and cholesterol prior to treatment. After therapy on the 30th day in dogs of the 4th group with the addition of Cobactan antibiotic therapy, blood counts partially restored. The amount of albumin remained low (22.2 ± 4.35 g / l), and AST increased to 56.08 ± 19.61 U / l. In dogs of the fifth group, a decrease in the amount of total protein and albumin remained. The sixth group retained a low content of total protein and albumin, and AST increased to 47.1 ± 9.9 U / L and alkaline phosphatase to 87.6 ± 12.41 U / L (Table 4).

Table 4: Blood biochemical parameters in dogs infested by Cystoisospora sp. during treatment, M ± m

Values	Units	Before therapy	30 days after therapy			Reference values
			4th group Stop Coccid + Cobactan 2.5% (n=9)	5th group Stop-Coccid + Pro-Colin (n=9)	6th group Stop-Coccid (n=6)	
Total bilirubin	μmol / l	1.9±2.51	7.71±2.21	2.2±0.58	4.6±1.04	< 13.5
Direct bilirubin	μmol / l	0.3±0.08	0.00±0.00	0.6±0.00	1.8±0.11	< 5.5
AST	U / L	63.0±4.97*	56.08± 19.61	29.0±8.63	47.1±9.95	over 6 months: 8-42 (up to 6 months: <70)
ALT	U / L	51.0±6.22	46.24±14.21	44.00±12.64	31.00±8.77	10 – 58
Ritis coefficient	units	1.0±0.23	1.21±0.26	1.2±0.26	1.2±0.26	1.1 – 1.3
Urea	mmol / l	2.90±0.40*	4.14±0.54	3.6±0.31	3.8±0.27	3.5 – 9.2
Creatinine	μmol / l	126.0± 16.21	98.17±17.24	116.0±16.75	121.0±15.22	54-138 (44-90 dogs up to 10 kg)
Total protein	g / l	46.0±3.73*	57.4±5.21	52.6±4.12	53.1±3.41	over 6 months: 55-73 (up to 6 months: 44-56)
Albumin	g / l	21.0±3.45	22.2±4.35	23.4±4.21	23.0±4.65	25 – 39
Alkaline phosphatase	U / L	53.0±11.36	47.8±13.41	38.2±12.64	87.6±12.41	over 8 months: 10-70 (up to 8 months: 80-230)
Alpha amylase, total	U / L	942.0± 208.37	1174.5± 236.54	959.0± 221.15	1004.0± 214.66	300 -1500 (over 4 months)
Glucose	mmol / l	5.3±0.93	4.65±0.92	5.8±0.93	5.4±0.92	3.3 – 6.3
Cholesterol	mmol / l	2.2±0.65*	3.33±0.84	3.1±0.72	3.6±0.54	2.5-6.0
Triglycerides	mmol / l	0.30±0.16	0.77±0.16	0.42±0.13	0.41±0.14	0.15-0.84
LDH	U / L	196.0± 41.68	147.3±32.07	183.0±41.85	164.0±31.34	23 – 220
Globulin	g / l	24.0±4.64	37.0±4.86	36.0±4.15	32.0±3.77	26 – 44

Note. Reference values are given by A.M. Barger and A.L. MacNeill (2015). P <0.05.
* - differences between the compared values are significant.

Compared with the pre-treatment indicators, biochemical parameters after giardiasis, and cystoisisporosis therapy with the use of the antibiotic Cobactan 2.5% came to reference values on the 30th day. Only albumin levels in dogs with cystoisosporosis have not fully recovered. This is due to the development of inflammation and destruction of the mucous membrane of the small intestine and impaired absorption of proteins from food. After the use of specific therapy and prebiotic on the 30th day, an incomplete recovery of the amount of blood proteins was noted, and in case of giardiasis, cholesterol. In groups without additional drugs with protein and cholesterol deficiency, the urea level did not recover either.

With prolonged follow-up of patients for 6 months in groups using an antibiotic in 6% of cases, gastrointestinal tract malfunctions were clinically manifested (a change in the consistency of feces towards softening, refusal to feed, and vomiting). No similar violations were observed in prebiotic groups.

Consequently, antibiotic therapy in case of giardiasis and cystoisosporosis allows the invasive dogs to recover faster against the background of specific therapy. But we assume that without the addition of a prebiotic, recovery with this scheme is incomplete [20, 41-50].

The second series of experiments consisted in comparing two complex treatment regimens: 1. the main antiprotozoal drug + antibiotic + probiotic and 2. the main antiprotozoal drug + antibiotic + probiotic + complex B vitamins (Milgamma, Worwag Pharma GmbH & Co KG (Germany), solution for intramuscular injection.

Formed 3 groups of dogs with giardiasis: 7th group - received therapy according to the scheme - Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin; 8th group - received therapy according to the scheme - Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma; 9th group - was treated only with Drontal plus antiprotozoal drug and was a control.

According to hematological indicators, animals of the 8th group with the addition of B vitamins to the treatment regimen had the best blood counts. In dogs of the 9th group, the indicators are at the lower boundary of the reference values and were the lowest when comparing the three groups (Table 5).

Table 5: Complete blood count in dogs infested by Giardia sp. during complex treatment, M ± m

Value	Before therapy	30 days after therapy			Reference values
		7th group Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin (n=6)	8th group Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma (n=6)	9th group (n=4)	
RBC, x10^{12}/л	5.7±0.81	6.5±1.8	7.9±2.3	6.0±1.9	5.5-8.5
WBC, x10^{9}/л	22.6±2.3	15.4±2.4*	7.3±2.1	8.6±3.0	6-17
HB, g / l	146.5±3.7	127.1±3.8	136.4±3.2	162.7±4.1	120-180
PLT, x10^{9}/л	343.4±2.4	468.1±3.8	413.6±3.9	485.4±4.2	200-900
HCT, %	39.4±1.7	38.5±1.8*	43.4±1.4*	41.3±1.7*	37-55
ESR, mm/h	4.4±1.4	2.7±1.6	3.4±1.6	3.7±1.4	2.0-3.5
MCV, fl	62.8±2.2	63.2±2.1*	61.3±2.0*	62.0±2.2*	60-75
MCHC, %	33.5±3.8	33.4±4.6*	32.7±4.0*	34.3±3.4*	32-36
RDW, %	13.9±2.0	15.3±2.0	14.1±2.3	13.7±2.1	11.9-16.0
MCH, pg	22.6±0.5	24.9±0.8	28.2±0.7	26.1±0.6	21-27
Leukogram					
Monocytes, %	6.0±0.02	7.0±0.03	6.0±0.3	7.0±0.02	3-10
Lymphocytes, %	23.0±1.7	17.0±2.0	11.0±1.3	14.0±1.3	12-30
Basophils, %	0	0	1	0	0-1
Eosinophils, %	3	4	6	7	2-10
Neutrophils:					
Band neutrophil, %	6.0±0.42	2.0±0.34	3.0±0.5	3.0±0.5	0-3
Segmented neutrophil, %	62.0±0.9	70.0±2.8	63.0±2.9	69.0±3.1	60-70

Note. Reference values are given by J.W. Harvey (2012)
P < 0,05.
* - differences between the compared values are significant

After treatment in dogs of the 7th group with therapy according to the main antiprotozoal drug + antibiotic + probiotic regimen, on the 30th day, blood counts recovered and reached reference values, except for albumin, which

remained at the lower boundary of the reference values (24.8 ± 4.21 g / l). In dogs of the 8th group, the amount of albumin reached a lower norm (25.4 ± 4.21 g / l). The remaining indicators reached compliance with reference values (Table 6).

Table 6: Blood biochemical parameters in dogs infested by Giardia sp. during complex treatment, M ± m

Values	Units	Before therapy	30 days after therapy			Reference values
			7th group Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin (n=6)	8th group Drontal plus + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma (n=6)	9th group (n=4)	
Total bilirubin	μmol / l	3.4±0.75	4.1±0.81	3.2±0.64	4.2±0.71	< 13.5
Direct bilirubin	μmol / l	2.1±0.03	1.2±0.02	1.6±0.02	1.7±0.01	< 5.5
AST	U / L	64.2±7.81	32.1±4.62	29.0±3.43	28.4±3.75*	over 6 months: 8-42 (up to 6 months: <70)
ALT	U / L	62.0±5.10	31.4±6.82	22.0±5.79*	27.0±6.24	10 – 58
Ritis coefficient	units	1.15	1.12	1.32	1.07	1.1 – 1.3
Urea	mmol / l	3.2±0.63	4.14±0.54	4.2±0.44*	3.8±0.42	3.5 – 9.2
Creatinine	μmol / l	124.0±14.43	93.17±15.22	117.0±13.77	112.0±14.25	54-138 (44-90 dogs up to 10 kg)
Total protein	g / l	44.0±4.81	56.4±5.12	59.0±4.16*	51.0±4.17	over 6 months: 55-73 (up to 6 months: 44-56)
Albumin	g / l	20.0±2.54	24.8±2.21	25.4±2.21	22.1±2.83	25 – 39
Alkaline phosphatase	U / L	51.0±23.35	48.87±23.34	38.0±22.66	43.0±21.45	over 8 months: 10-70 (up to 8 months: 80-230)
Alpha amylase, total	U / L	871.3± 144.58	1104.5± 256.52	915.0± 231.85	986.0± 231.57	300 -1500 (over 4 months)
Glucose	mmol / l	5.41±0.94	5.65±0.92	5.3±0.93	5.68±0.91	3.3 – 6.3
Cholesterol	mmol / l	2.1±0.17	3.6±0.13*	2.5±0.16	2.4±0.14	2.5-6.0
Triglycerides	mmol / l	0.44±0.13	0.77±0.14	0.42±0.11	0.56±0.18	0.15-0.84
LDH	U / L	192.0± 42.31	247.38± 53.04	186.0± 41.68	216.0±43.80	23 – 220
Globulin	g / l	23.0±3.834	31.0±4.8	27.0±4.85	25.0±3.22	26 – 44

Note. Reference values are given by A.M. Barger and A.L. MacNeill (2015). P <0.05.
* - differences between the compared values are significant.

Dogs with cystoisosporosis were divided into 3 groups: the 10th group received the following therapy: Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin; The 11th group received the Stop-Coccid + Cobactan 2.5% scheme + Pro-Colin prebiotic + Milgamma; The 12th group was treated only with Stop- Coccid and was a control. Hematological research indices reached the level of reference values on the 30th day after the specific therapy (Table 7).

Table 7: Changes in complete blood count in dogs infested by Cystoisospora sp. during complex treatment, M ± m

Value	Cystoisospora sp. invasion	30 days after therapy			Reference values
		10th group Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin (n=5)	11th group Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma (n=5)	12th group Stop- Coccid (n=4)	
RBC, $x10^{12}$/л	6.9±1.1	6.3±1.8	8.1±1.6	6.7±1.9	5.5-8.5
WBC, $x10^{9}$/л	26.2±2.9*	14.6±2.0	14.9±2.4	11.8±2.7	6-17
HB, g / l	153.2±3.5	146.2±3.9	168.3±4.6	131.4±4.5	120-180

PLT, x10^9/л	438.7±3.4	529.4±6.3	460.5±5.5	501.1±5.4	200-900
HCT, %	42.9±1.2	44.8±3.5	41.3±4.1	45.5±3.7	37-55
ESR, mm/h	3.1±1.2	2.6±0.8	2.9±0.7	2.4±0.8	2.0-3.5
MCV, fl	64.0±2.7	67.2±2.9	66.7±2.4	66.0±2.8	60-75
MCHC, %	31.9±4.6	34.2±5.1	32.4±4.0	35.1±4.2	32-36
RDW, %	15.1±2.1	13.6±3.1	14.8±2.9	15.6±3.0	11.9-16.0
MCH, pg	24.0±0.7	23.1±1.0	25.1±0.8	23.8±0.8	21-27
Monocytes, %	8.0±0.3	9.0±0.2	8.0±0.3	6.0±0.1	3-10
Lymphocytes, %	26.0±1.9	24.0±2.1	17.0±1.9	22.0±1.7	12-30
Basophils, %	1	0	1	0	0-1
Eosinophils, %	5*	2	4	2	2-10
Neutrophils:					
Band neutrophil, %	4.0±0.3	2.0±0.1	2.0±0.1	3.0±0.2	0-3
Segmented neutrophil, %	66.0±1.6	63.0±4.0	68.0±4.1	67.0±3.9	60-70

Note. Reference values are given by J.W. Harvey (2012)
P < 0,05.
* - differences between the compared values are significant.

Dogs of the 10th group with confirmed cystoisosporosis after therapy on the 30th day had a low level of albumin (23.17 ± 4.35 g / l). In dogs of the 11th group, all indicators reached normal values for the species (Table 8).

Table 8: Blood biochemical parameters in dogs infested by Cystoisospora sp. during complex treatment, M ± m

Values	Units	Before therapy	30 days after therapy			Reference values
			10th group Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin (n=5)	11th group Stop-Coccid + Cobactan 2.5% + prebiotic Pro-Colin + Milgamma (n=5)	12th group Stop-Coccid (n=4)	
Total bilirubin	μmol / l	1.9±0.41	7.71±1.51	2.2±0.38	4.6±0.34	< 13.5
Direct bilirubin	μmol / l	0.3±0.08	0.00±0.00	0.6±0.00	0.8±0.11	< 5.5
AST	U / L	53.0±9.43	38.1±9.65	29.0±8.61	47.1±9.90	over 6 months: 8-42 (up to 6 months: <70)
ALT	U / L	71.0±6.22	46.24±5.24	44.0±6.64	31±5.77*	10 – 58
Ritis coefficient	units	1.0±0.23	1.21±0.26	1.2±0.26	1.2±0.26	1.1 – 1.3
Urea	mmol / l	2.9±0.36	4.14±0.54*	3.6±0.34	3.8±0.27	3.5 – 9.2
Creatinine	μmol / l	126.0± 16.21	98.17±17.24	116.0±16.75	121.0±15.26	54-138 (44-90 dogs up to 10 kg)
Total protein	g / l	46.0±3.73	57.4±5.23	58.1±4.34	53.1±3.41*	over 6 months: 55-73 (up to 6 months: 44-56)
Albumin	g / l	21.0±3.45	23.17±4.35	26.3±4.17	23.0±4.65	25 – 39
Alkaline phosphatase	U / L	53.0±6.15	47.8±7.44	38.2±5.60	87.6±7.44	over 8 months: 10-70 (up to 8 months: 80-230)
Alpha amylase, total	U / L	942.0± 188.33	1174.5± 216.54	959.0± 201.15	1004.0± 214.67	300 -1500 (over 4 months)
Glucose	mmol / l	5.3±0.93	4.65±0.92	5.8±0.93	5.4±0.92	3.3 – 6.3
Cholesterol	mmol / l	2.2±0.45	3.33±0.54	3.1±0.70	3.6±0.58*	2.5-6.0
Triglycerides	mmol / l	0.30±0.11	0.77±0.16	0.42±0.13	0.41±0.14	0.15-0.84
LDH	U / L	196.0± 41.68	177.3±42.02*	183.0±31.84*	174.0±41.35*	23 – 220
Globulin	g / l	24.0±4.64	37.0±4.87	36.0±4.10	32.0±3.73	26 – 44

Note. Reference values are given by A.M. Barger and A.L. MacNeill (2015). P <0.05.
* - differences between the compared values are significant.

During long-term follow-up of patients for 6 months in the experimental groups with both the first and second treatment regimens, cases of gastrointestinal tract functioning were clinically manifested (change in stool consistency towards softening, refusal to feed, vomiting). Consequently, antibiotic therapy with the addition of a probiotic in case of giardiasis and cystoisosporosis allows more fully recovering invaded dogs on the background of specific therapy [20, 21, 22].

Treatment for malabsorption includes changing the diet, treating complications, and addressing the cause if it can be identified. Effective treatment of small bowel disease depends on the nature of the disorder, but when a specific diagnosis cannot be made, treatment can be provided on an experimental basis. The prognosis worsens for dogs with severe small bowel disease, cancer, fluid retention caused by low protein levels, severe weight loss, low levels of vitamin B12 in the blood, or lack of appetite (Merck & Co., 2020).

The prognosis in cases of malabsorption is good if there is a simple solution. The prognosis is worse, the heavier the pathology of the small intestine. A poor prognosis is associated with severe intestinal inflammation, tumor diseases, severe weight loss, hypoalbuminemia and ascites, anorexia and hypocobalaminemia [23, 24].

With giardiasis, fenbendazole (50 mg / kg / day for 2-5 days) or metronidazole (25 mg / kg / day for 5-7 days) are the most commonly used and are particularly effective. When diarrhea persists after a course of treatment, repeated tests and the appointment of alternative drugs are necessary [10].

According to the data of A. Montoya (2008), in animals treated with Drontal® plus, the clinical signs disappeared and cyst production ceased, in comparison with the control group that did not receive the drug (Montoya A., 2008). It includes pyrantel embonate, praziquantel, febantel. But there is a study in which re-isolation of cysts was noted on the 7th day after treatment [25, 26, 54-60].

In 2015, Konyaev S.V. et al noted in the treatment of giardiasis with Drontal® plus the disappearance of clinical signs. After treatment, all dogs had a negative control test for the Giardia antigen. In two animals, the frequency of bowel movements decreased, and diarrhea persisted, despite the treatment. Further clinical and laboratory research established a combined invasion with cystoisospores in them [27, 61-66]. As shown in Payne P.A et al. (2002) when treating animals with giardiasis, it is very important to observe sanitary rules and exclude the possibility of reinfestation [28, 34-41].

In our work, the therapy of giardiasis in domestic dogs with Drontal plus reinvasion for 6 months was not observed.

In cystoisosporosis, trimethoprim-sulfadiazine or other sulfa drugs are usually used. For example, sulfadiamethoxin. The most effective are considered to be ponazuril or doltrazuril for dogs. Changing the diet, environmental analysis and treatment of all animals in contact is also important for cystoisosporosis [10, 31-36].

Diet therapy for ulcers and inflammation of the stomach and duodenum is the basis of any treatment regimen in dogs. The feed product should be non-greasy, easily digestible, well eaten by the pet and not irritate the mucous membrane. Purina EN dietary food fully complies with the listed requirements. In addition, due to the high content of medium chain triglycerides and the restriction of long chain (less than 10% dry matter), it provides functional unloading of the pancreas. The number of enzymes, primarily lipase, secreted into the intestinal lumen decreases, therefore, the intensity of the aggressive chemical effect on the mucosa decreases. Dietary food EN contributes to the restoration of the intestinal mucosa, its motility and secretory function [29, 30].

CONCLUSION

When treating dogs with giardiasis and cystoisosporosis, the main antiprotozoal drug (Drontal for giardiasis or Stop-Coccid for cystoisosporosis) + antibiotic Cobactan + Probiotic Pro-Colin + vitamin B complex (Milgamma), the dogs were restored 100% on the 30th day after the end of treatment. During long-term follow-up of patients with clinical manifestations of gastrointestinal tract malfunction (a change in the consistency of feces towards softening, refusal to feed, and vomiting) with the use of a prebiotic was not observed.

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CONFLICT OF INTEREST

The authors have no conflict of interest.

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