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Hemogram of buffaloes (*bubalus bubalis*) wormed and nonwormed by helminthes

Hemograma de búfalos (*bubalus bubalis*) parasitados e não parasitados por helmintos

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Abstract

The gastrointestinal parasitoses of domestic animals have a great impact on the hematological status but nonetheless research on this subject concerning buffaloes is rare. Thus, the blood profile of buffalo calves naturally infected by parasites from the seventh to the 300th day of life. Animals were distributed in three experimental groups treated with ivermectin, fenbendazole, and placebo, respectively. Changes of hematological parameters were related to the presence of adult gastrointestinal nematodes, which were diagnosed and identified by the presence of eggs in the feces of wormed animals as compared to those medicated.

Keywords: buffalo, helminthes, hemogram, ivermectin, fenbendazole.

Resumo

É reconhecida a capacidade das parasitoses gastrintestinais em alterar os índices hematológicos dos animais, no entato não está descrita na literatura de forma detalhada este fenômeno em búfalos. Para isto foi realizado o estudo do perfil hematológico de bezerros bubalinos naturalmente infectados por parasitos gastrintestinais do 7º ao 300º dia de vida. Os animais formaram três grupos experimentais tratados com ivermectina, fenbendazole e placebo.

Foram encontradas alterações no quadro hematológico concomitante com a presença de nematoides adultos parasitando os animais, que tiveram sua presença confirmada pelo diagnóstico de ovos nas fezes dos animais medicados e não medicados.

Palavras-chave: búfalo, helmintos, hematologia, ivermectina, fenbendazole.

Introduction

Helminthiases are considered limiting factors in buffalo production once they are responsible for the low development in young cattle and for mortality increase. The presence of parasites in the gastrointestinal tract significantly interferes with the production and activation processes of the hemopoietic tissue leading to anemia, hypoproteinemia, decrease of immunoglobulin levels, and leukocytosis. These changes found in wormed animals may occur by activation of the immune response, intestinal hemorrhage, and impairment of digestion and absorption of nutrients caused by the spoliative action of helminthes.

Dargie e Allonby (1975) demonstrated that anemia in Merino sheep resulted from severe hemonchosis developed in three stages. In the first one, from the seventh to the twenty-fifth day after infection, anemia resulted from direct loss of blood; the erythropoietic response was absent, and the serum iron concentration was normal. In the second stage, from the sixth to the fourteenth week after infection, a regenerative anemia persisted due to the increase of erythropoiesis resulting from the ongoing blood loss. During this phase, the absorption and the availability of iron by the organism began to decrease, and its concentration increased in feces. The third stage is noteworthy for the severity of the anemia status resulting from the decrease of erythropoiesis due to the unavailabily of iron in blood and in bone marrow, which was related to a possible decrease in the globin synthesis, aminoacid with will result in reduced production in the proteic digestion disorder. The progressive hemonchosis culminated in nonregenerative anemia resulting from iron defiency.

Nonhematophagous helminthes such as *Trichostrongylus* spp. may also be involved with the anemia development in chronical infections. These act by the inhibition of erythropoiesis by means of competition for nutrients and interference with digestion, absorption, and availability of food nutrients such as copper, iron, and specific amino acids (Jain, 1986).

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Láu (1985) assessed the hematological profile of buffalo calves naturally wormed by *Neoascaris vitulorum* and found a significant decrease of the number of erythrocytes, as well as reduction of the concentration of hemoglobin and of the hematocrit. The parasited animals also presented leukocytosis with eosinophilia.

Patel et al. (1971) reported an increased number of erythrocytes and of the concentration of hemoglobin in healthy bulls during the summer, indicating a seasonal variation. Nevertheless, seasonal interference was not observed in calves once significant increases in the erythrocytes total count, in the concentration of hemoglobin, and in the leukocytes total count were recorded, especially in eosinophils and lymphocytes, in all seasons of the year.

As for age, Santos (1984) studied the hematological parameters of calves naturally infested infected with nematodes and concluded that the total number of leukocytes was smaller from the first to the 15th day of life, with considerable posterior increase until weaning (180 days of life). The neutrophil: lymphocyte ratio in the interval from the first to the fifteenth day of life was 1.29 in male buffaloes and 1.02 in females; nonetheless, the same relation ratio verified on the 180th day was 0.28 in males and 0.24 in females.

Silva et al. (1992) studied the erithrogram of three healthy buffalo breeds belonging to herds located at the Vale do Ribeira, in the State of São Paulo, Brazil. The findings concerning the hematological values did not present any variation among the breeds, but significantly varied in relation to the age of the animal at the time of collection. With the aging of the animals, the values of erythrocytes hemoglobin (Hb) and hematocrit (HCT) decreased, while the hematometric indexes such as mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) increased.

After analyzing hemograms of Murrah buffaloes, from birth to one year of age, Bonfim (1995) reported of erythrocytes, of Hb, and of the HCT after 90 days. This decrease was ascribed to the size diminishing of erythrocytes in the first three months, followed by a gradual increase and a consequent decrease of the number. This study concluded that the age factor significantly influenced the values of erythrocytes, Hb, HCT, MCH, MCHC, total leukocytes, segmented neutrophils, lymphocytes, eosinophils, and basophils.

As no other work reported the hematological status of buffalo calves wormed by helminthes and of wormed calves up to 300 days of life, this study aimed at evaluating the influence of gastrointestinal parasites on the hematological status of them.

Material and methods

The study was made with a group of animals raised at the F*azenda Canoas and Fazenda Canoas do Jorge Grande*, respectively identified as ranches A and B, located at Dores do Indaiá county, in the State of Minas Gerais, Brazil.

The animals were raised under the semi-intensive system. Napier grass and corn bran supplementation was provided for lactating buffalo females during the winter and for calves throughout the year. In ranch A calves were fed a liter of milk less per day as compared to Ranch B calves. The properties were neighbours, on slightly irregular terrain, mixed pastures with native roughage and *Brachiaria decumbens,* and had similar facilities. Buffaloes and cattle were simultaneously raised, without any separation per age in both properties.

This study followed 53 Jafarabadi buffaloes ranging from 7 to 300 days of age every 30 days for 24 months. Out of 53, 23 calves belonged to Ranch A, 11 being males and 12 females, and 30 calves belonged to Ranch B, being 10 males and 20 females. Both cattle and buffalo calves shared the same environment and were only set apart at milking times. Ranch A and Ranch B animals were randomly distributed in six different groups according to birth order.

The animals belonging to Ranch A composed groups (G) 1, 2, and 3 with five, seven, and eleven individuals, respectively. G1 animals were dewormed on the seventh day of life and subsequently every 60 days with 200 microgramas (mcg) of ivermectin per kilogramg of body weigh, subcutaneously; G2 animals were dewormed on the 15th, 30th, 60th and 180th days with 10mg of fenbendazole per kilogramg kg of body weigh, orally; G3 animals were not dewormed and served as control group.

The animals belonging to Ranch B composed groups (G) 4, 5, and 6 with ten, eleven, and nine individuals, respectively. G4 animals were dewormed on the 7th day of life and subsequently every 60 days with 200mcg of ivermectin per kg of body weigh, subcutaneously; G5 animals were dewormed on the 15th, 30th, 60th and 180th days with 10mg of fenbendazole per kg of body weigh, orally; G6 animals were not dewormed and served as control group, breed in the same conditions of Ranch A.

The calves were monthly and individually corralled for feces and blood collection and for weight assessment with appropriate metric tape. Animals born during collection intervals were included in groups according to their age and study proposal for each group.

Aiming at the diagnosis of helminthes in order to correlate contingent differences in the hemogram of animals belonging to different groups, egg per gram of feces counts (EPG) were made in McMaster chamber, according to Gordon & Whitlock (1939), modified by Whitlock (1948) according to Ueno & Gonçalves (1998), besides coprocultures for the identification of helminthes genera present in the feces samples positive to the EPG exam, according to O´Sullivan´s (1950) methodology described by Ueno & Gonçalves (1998). The nematode larvae were collected by the Baermann´s technique, mentioned by Ueno &Gonçalves (1998).

Blood collection was made by punction of the jugular vein, in 5mL glass flasks containing the anticoagulant 10% Ethylenediaminetetraacetic Acid (EDTA) for the total count of erythrocytes and leukocytes; determinations of the hemoglobin concentration (Hb), of the hematocrit (HCT), and of the hemacytometric indexes (MCV, MCH, and MCHC) in electronic instrument[1] using the impedance method to classify the different cell types. The erythrocytes diameter is used by the device as reference for the identification of the cell types

1 Blood Counter HORIBA ABX Diagnostics, Califórnia - USA

of different animal species. The erythrocytes diameter of buffaloes is similar to that of cattle allowing the configuration of the mentioned instrument to be applied to bovines upon examination (Jain 1986). The total plasma protein concentration was determined by the colorimetric test *in vitro* by biuret according to Kaneko et al. (2008).

Blood smears were made prior to the hemogram and were stained with May- Grünwald-Giemsa for subsequent differential count of leukocytes in an optical microscope, according to Jain (1986).

For the identification of the helminthes species, a necropsy of a Group 6 animal was made. Feces samples were collected in all segments of the gastrointestinal treat and fixed in 10% formol solution.

The mean values were submitted to the statistical analysis using the SNK test with 5% of significance with the aid of Softwear S.A.S. 1998 version.

Toxocara spp*., Strongyloides* spp*., Haemonchus* spp*., Cooperia* spp*., Paracooperia spp*., and *Oesophagostomun* spp. were identified by the microscopical features of the eggs and larvae present in the feces in accordance with Levine (1980).

Results

Toxocara vitulorum, *Strongyloides papillosus*, *Haemonchus similis*, *Haemonchus contortus*, *Ostertagia trifurcata*, *Cooperia punctata*, *Paracooperia nodulosa*, and *Oesophagostomun radiatum* were identified in the feces of the necropsed animals. Lesions in the mucosa of the small intestine and caecum and hypertrophia of mesenteric lymphonodes were visualized. The histopathological study of the transversal cut of intestinal node showed the presence of *P. nodulosa* larvae with intense accumulation of epiteloid macrophages, lymphocytes, and multifocal infiltration of eosinophils (Bastianetto et al., 2005). After interruption of medication of the animals on the 180th day of life in Ranch A, G2 animals presented an EPG increase in feces and decreased number of erythrocytes in the circulating blood (Tables 1 and 4).

Table 1: Mean count of helminth eggs number per gram of feces of buffalo calves belonging to two properties located in Dores do Indaiá county in the State of Minas Gerais, Brazil, dewormed with 200mcg of ivermectin per kg (G1 and G4), dewormed with 10mg of fenbendazole per kg (G2 and G5), and not dewormed (G3 and G6)

Age (days)	G1		G2		G3		G4		G5		G6	
	N	EPG	N	EPG	N	EPG	N	EPG	N	EPG	N	EPG
7	2	0	0	0	4	0	2	100±141.42^{a}	3	1000.00±1732.05^{a}	3	33.33±57.73^{a}
15	2	0	2	0	3	933.33±1616.58^{b}	3	0	4	0	3	0
30	2	0	4	12750±602.07^{a}	4	2000±2423.49^{b}	3	0	5	0	5	180±349.28^{b}
60	4	25±50^{a}	7	125±353.55^{b}	1	3346.15±7621.95^{b}	9	700±2100^{b}	11	0	8	5437.50±10789.93^{c}
90	5	33.33±81.64^{b}	7	71.42±111.26^{a}	11	1045.45±1407.38^{b}	9	0	11	72.70±210.19^{b}	9	344.44±804.84^{a}
120	5	40±54.77^{a}	7	157.14±214.91^{a}	9	533.33±800^{b}	9	0	11	0	8	75±116.49^{a}
150	4	25±50^{a}	7	42.85±113.38^{b}	10	100±105.40^{c}	9	0	9	0	9	11.11±33.33^{e}
180	5	0	6	100±126.49^{a}	10	40±96.60^{d}	8	12.50±35.35^{b}	11	0	9	10±31.62^{e}
210	4	0	7	14.28±37.79^{b}	10	70±105.93^{b}	10	0	11	0	8	0
240	5	0	7	0	9	44.44±52.70^{b}	9	0	9	0	7	0
270	5	20±44.72^{a}	4	50±100^{c}	10	60±134.98^{e}	7	0	11	0	9	190.90±326.96^{b}
300	2	80±109.54^{c}	7	0	6	16.66±40.82^{f}	9	11.11±33.33^{b}	1	75.00±200.56^{b}	9	55.55±133.33^{a}

Mean e followed by distinct letters in the same column are different (p<0.05)

The mean values of the analyzed parametres of animals composing G1 to G6 are demonstrated in Tables 2 to 7. A significant difference (P<0.01) among the mean values of erythrocytes, Hb, HCT, and MCH was found as compared to the means of these parameters between G2 and G4 and G1 and G5 animals. A significant difference (P<0.01) in relation to MCV in animals of different genders was also observed.

Due to the antiparasitic effect of the medicine and the consequent absence or decrease of the spoliation caused by the helminth, the decrease of the erythrocyte number in G1 and G4 animals during the experimental period was not observed (Tables 2 and 3). At 240 days of age, all animals presented similar mean counts of erythrocytes. The mean values of the hematological indexes found in all animals are presented (Tables 2 to 7) according to the groups.

The hematological parameters analysis, regardless the raising system, shows that G3 and G6 animals presented reduced concentration of Hb and MCH. A significant difference (P<0.01) among the mean values of erythrocytes, Hb, HCT, and MCH was found as compared to the means of these parameters between G2 and G4 and G1 and G5 animals.

In the differential count of leukocytes (Tables 8, 9, 10, 11, 12, and 13), a significant difference (p<0.01) was found in relation to the month of collection in terms of number of lymphocytes, monocytes, segmented neutrophils, stab neutrophils, and eosinophils.

The number of eosinophils presented a significant difference (p<0.01) as compared to the parameters: raising system, month of collection, and interaction of raising system, month of collection, and treatment factors. G6 animals presented a greater number of eosinophils reaching $6.33 \pm 11.01 \times 10^3$/mL on the 60th day and $6.22 \pm 5.51 \times 10^3$/mL on the 240th day of age (Table 12), older animals normally expected to have high parasitic infestation.

Discussion

Hematophagous and nonhematophagous parasites may cause anemia in wormed animals. Apparently, the bleeding caused by some helminthes species is the primary factor in the triggering of the anemic status as in the infection by

Table 2: Mean values and standard deviation of hematological indexes of Jafarabadi race buffaloes from the seventh to the 300th day of life, raised at Ranch A, dewormed on the seventh day of life and subsequently every 60 days with 200mcg of ivermectin per kg, subcutaneously

Groupo	Age (days)	Nº obs	Erythrocytes ($X10^6/mL^3$)	Hb (g/dL)	MCV (fL)	MCH (Pg)	MCHC (g/dL)	HCT (%)	TP (g/dL)
G1	7	2	9.09±0.18	12.15±1.48	42.50±2.12	14.10	33.30±1.41	38.35±0.91	8.93±0.04
G1	15	4	8.63±0.48	11.15±0.91	42.50±0.70	14.00±0.42	32.95±0.07	36.55±0.91	8.70±2.26
G1	30	4	7.32±1.30	12.50±1.16	39.50±4.94	14.10±0.98	35.80±2.54	29.25±9.26	9.01±1.28
G1	60	11	8.99±1.02	12.08±0.94	40.00±2.16	12.95±0.78	32.05±0.95	36.30±4.90	7.10±0.28
G1	90	10	9.93±1.04	11.88±1.24	38.50±2.38	13.50±0.67	34.85±0.86	37.70±2.63	9.11±1.99
G1	120	8	8.92±1.46	12.25±0.68	40.60±1.51	13.02±0.46	32.02±1.13	36.18±5.65	7.59±0.43
G1	150	1	7.87	10.87±0.60	40	13.40	33.60	31.30	7.75
G1	180	7	7.79±1.25	10.49±1.30	41.20±4.32	13.68±1.84	33.06±1.05	31.90±2.59	7.51±2.18
G1	210	11	8.55±1.49	10.50±0.72	40.00±4.24	13.22±2.01	32.70±1.83	34.02±2.91	7.05±0.83
G1	240	12	8.72±1.22	10.13±2.71	41.66±3.82	13.01±1.88	31.16±2.18	36.00±2.64	6.27±0.94
G1	270	9	8.62±0.82	11.50±0.98	42.20±1.64	13.38±0.85	31.80±1.06	36.28±4.43	6.21±0.89
G1	300	11	8.28±0.73	11.23±0.48	43.40±1.67	13.78±0.80	31.82±1.54	35.78±2.77	5.76±0.22

Table 3: Mean values and standard deviation of the hematological indexes of Jafarabadi buffaloes from the seventh to the 300th day of life, raised at Ranch B, dewormed on the seventh day of life and subsequently every 60 days with 200mcg of ivermectin per kg, subcutaneously

Group	Age (days)	Nº obs	Erythrocytes ($X 10^6/mm^3$)	Hb (g/dl)	MCV (Fl)	MCH (Pg)	MCHC (g/dl)	HCT (%)	TP (g/dl)
G4	15	1	7.16	10.80	44	15	33.80	31.80	11.20
G4	30	3	8.67±0.59	12.20±1.17	44.33±2.08	14.06±1.06	31.63±1.66	38.53±2.20	7.10
G4	60	3	9.03±1.61	11.90±2.26	41.66±0.57	13.16±0.23	31.56±0.87	37.56±6.37	8.67±1.12
G4	90	9	10.10±0.95	12.86±0.76	39.44±3.46	12.82±1.04	32.52±1.94	39.61±2.34	9.03±1.15
G4	120	8	10.30±1.39	12.77±1.55	39.25±3.49	12.50±1.18	31.97±2.42	39.96±3.43	7.28±1.51
G4	150	6	10.32±2.07	12.68±1.34	39.33±4.54	12.48±1.24	31.60±1.73	35.21±13.30	8.53±2.45
G4	180	8	9.16±1.00	11.60±0.48	39.75±3.19	12.75±1.09	32.08±1.49	36.18±1.86	7.19±0.71
G4	210	8	8.35±1.01	10.57±0.80	40.87±3.68	12.85±1.49	31.48±1.62	33.60±2.69	6.95±0.74
G4	240	11	8.76±0.84	11.10±1.02	40.63±2.97	12.72±1.12	31.27±1.84	35.56±2.96	6.22±0.87
G4	270	8	8.67±0.91	10.42±1.61	41.37±3.37	12.13±1.97	29.23±3.97	35.75±3.41	6.56±1.41
G4	300	9	9.17±1.02	11.78±1.04	41.77±3.70	12.92±1.24	30.98±0.90	38.00±2.76	6.03±0.56

Table 4: Mean values and standard deviation of the hematological indexes of pure and half-breed Jafarabadi buffaloes on the fifteenth and 300th day of life, raised at Ranch A, dewormed on the 15th, 30th, 60th, and 180th days of life, with 10mg of fenbendazole per kg of live weight, orally

Group	Age (days)	Nº obs	Erythrocytes ($X10^6/mm^3$)	Hb (g/dl)	MCV (Fl)	MCH (Pg)	MCHC (g/dl)	HCT (%)	TP (g/dl)
G2	15	2	7.83±0.35	11.95±0.21	44.50±2.12	15.25±0.91	34.10±0.28	35.00±0.28	8.10±0.14
G2	30	4	6.54±0.57	9.57±0.71	44.25±2.50	14.65±0.36	33.35±2.46	28.75±2.93	7.05±3.04
G2	60	7	7.80±1.84	10.52±2.60	39.85±3.02	13.48±0.88	34.08±1.14	30.97±7.80	7.96±2.02
G2	90	7	8.46±0.57	11.37±0.68	40.28±2.05	13.47±0.88	33.48±2.32	34.05±2.78	8.81±1.48
G2	120	4	8.37±0.69	11.30±0.51	42.25±0.50	13.57±0.84	31.85±1.83	35.60±2.82	9.91±1.00
G2	150	4	8.09±0.67	10.82±0.73	41.75±2.75	13.42±1.12	32.05±1.31	33.65±1.57	6.82±1.05
G2	180	4	7.09±0.39	10.36±0.29	44.33±1.36	14.65±0.58	33.00±1.14	31.48±1.42	7.36±1.03
G2	210	6	7.56±1.11	10.17±0.43	42.71±2.92	13.67±1.73	31.90±2.30	32.11±3.08	6.84±1.20
G2	240	7	8.28±0.48	11.01±0.98	43.16±1.60	13.28±1.01	30.90±1.60	35.66±2.48	6.14±0.27
G2	270	3	7.96±0.55	11.20±0.40	44.33±1.15	14.10±0.52	31.96±1.26	35.16±2.56	6.11±0.66
G2	300	7	6.89±0.60	10.20±0.45	46.00±1.82	14.87±0.88	32.35±0.93	31.58±1.99	5.53±0.22

Table 5: Mean values and standard deviation of hematological indexes of pure and half-breed Jafarabadi buffaloes from the seventh to the 300th day of life, raised at Ranch B, dewormed on 15th, 30th, 60th and 180th day of life with 10mg of fenbendazole per kg of live weight, orally

Group	Age (days)	Nº obs	Erythrocytes ($X10^6/mm^3$)	Hb(g/dl)	MCV (Fl)	MCH (Pg)	MCHC (g/dl)	HCT (%)	TP (g/dl)
G5	15	2	8.33±1.08	12.15±1.48	46.50±0.70	14.65±0.21	31.65±0.07	38.50±4.66	8.20±0.14
G5	30	4	8.03±0.67	11.15±0.91	44.25±1.70	13.92±0.96	31.35±1.39	35.52±2.05	11.23±0.15
G5	60	4	9.42±0.43	12.50±1.16	40.25±1.25	13.27±0.60	32.95±1.39	38.00±2.61	8.49±1.19
G5	90	11	9.50±0.96	12.08±0.94	40.00±2.72	12.79±0.60	32.13±1.70	37.60±2.74	9.29±1.36
G5	120	10	9.65±1.12	11.88±1.24	39.87±2.29	12.31±0.65	30.92±1.78	38.36±3.09	8.00±2.06
G5	150	8	9.41±0.79	12.25±0.68	41.83±1.72	13.06±0.75	31.16±1.19	39.30±1.82	6.73±0.90
G5	180	6	8.00±1.16	10.87±0.60	41.42±2.69	13.02±1.14	31.42±1.14	34.75±2.87	7.56±1.00
G5	210	7	7.85±1.29	10.49±1.30	42.45±2.16	13.47±1.14	31.63±1.91	33.31±5.18	6.79±0.97
G5	240	11	8.30±0.66	10.50±0.72	42.33±1.96	12.71±1.10	29.18±3.39	35.02±2.39	6.31±1.07
G5	270	12	9.76±3.06	10.13±2.71	42.66±1.11	10.76±2.89	25.12±6.43	37.13±2.88	5.90±0.31
G5	300	9	8.77±0.83	11.50±0.98	42.72±1.10	13.10±0.41	30.81±0.79	37.41±3.63	5.84±0.78

Table 6: Mean values and standard deviation of hematological indexes and of total protein (TP) of pure and half-breed Jafarabadi buffaloes, raised in Ranch A, wormed from the seventh to the 300th day of life

Group	Age (days)	Nº obs	Erythrocytes ($X10^6/mm^3$)	Hb (g/dl)	MCV (Fl)	MCH (Pg)	MCHC (g/dl)	HCT (%)	TP (g/dl)
G3	7	4	7.38±0.91	10.45±1.47	44.75±1.25	14.12±0.51	31.55±1.79	33.10±4.52	5.00±1,00
G3	15	2	8.80±0.64	11.75±1.34	38.50±2.12	13.35±0.63	34.65±0.63	33.95±4.59	11.70±0.51
G3	30	4	6.42±0.89	9.02±1.26	41.25±2.62	14.05±0.36	34.05±2.24	26.47±2.49	7.74±0.53
G3	60	9	8.67±1.10	11.51±1.78	40.00±4.03	13.26±1.05	33.26±2.31	34.76±5.47	8.15±2.29
G3	90	7	7.81±1.93	10.40±2.27	40.00±3.60	13.45±1.24	33.71±1.37	30.84±6.14	7.25±1.56
G3	120	7	8.01±0.92	10.90±1.24	41.85±2.91	13.62±0.85	32.70±0.96	33.41±4.60	6.82±1.01
G3	150	7	8.17±0.97	10.37±1.00	40.00±2.51	12.72±1.34	31.80±1.83	32.57±2.87	7.25±2.29
G3	180	8	8.61±0.71	10.88±0.66	41.12±3.09	12.72±1.38	30.95±2.23	35.26±1.79	6.59±0.90
G3	210	10	8.44±0.99	11.28±0.53	41.40±2.98	13.46±1.38	32.41±1.31	34.80±2.18	6.77±0.73
G3	240	9	8.33±0.70	10.81±0.60	41.66±2.12	13.03±0.90	31.35±1.53	34.58±2.56	5.71±0.56
G3	270	10	8.36±0.64	11.39±0.63	42.80±1.93	13.65±0.77	31.98±0.77	35.62±2.13	5.80±0.48
G3	300	6	8.42±0.70	11.68±1.20	43.83±2.63	13.91±1.03	31.78±1.03	36.75±3.11	5.54±0.27

Table 7: Mean values and standard deviation of hematological indexes and total protein (TP) of pure and half-breed Jafarabadi buffaloes, raised in Ranch B, wormed from the seventh to the 300th day of life

Group	Age (days)	Nº obs	Erythrocytes ($X10^6/mm^3$)	Hb (g/dl)	MCV (Fl)	MCH (Pg)	MCHC (g/dl)	HCT (%)	TP (g/dl)
G6	7	12	7.85±1.16	10.30±2.08	39.33±2.08	13.00±0.81	32.96±0.68	31.13±5.85	10.90±0.98
G6	15	3	8.82±0.43	13.10±0.14	43.50±0.70	14.80±0.56	34.00±0.70	38.45±1.20	9.15±2.33
G6	30	2	8.06±0.45	10.18±0.75	38.80±2.58	12.62±0.68	32.50±1.86	31.34±2.51	9.48±1.15
G6	60	5	9.28±1.09	11.54±1.41	39.42±1.90	12.42±0.17	31.47±1.09	36.68±4.42	8.37±1.40
G6	90	7	9.82±0.96	11.50±1.13	36.25±2.62	11.72±0.26	32.52±2.58	35.45±3.48	9.29±0.96
G6	120	4	9.01±1.01	11.27±0.89	40.57±3.04	12.58±1.14	31.00±1.18	36.41±3.54	7.47±0.71
G6	150	7	8.70±1.23	11.15±1.08	40.00±3.22	12.88±1.12	32.20±0.72	34.60±3.17	6.78±1.95
G6	180	6	8.33±0.71	10.14±0.60	3.70±2.05	12.22±0.90	30.86±1.88	32.96±3.08	6.41±0.59
G6	210	10	8.30±0.81	10.78±1.18	41.00±2.56	13.01±0.95	31.81±1.61	33.90±2.84	6.63±0.90
G6	240	8	9.09±0.70	8.35±3.73	40.00±1.54	9.33±3.99	23.30±9.93	35.60±2.49	6.37±0.45
G6	270	6	8.50±0.43	11.28±0.46	42.54±2.33	13.30±0.67	34.06±9.29	36.15±1.38	6.22±0.58
G6	300	11	8.51±0.58	11.26±0.75	43.00±1.73	13.23±0.55	30.83±1.02	36.53±2.09	5.58±1.02

Table 8: Mean values and standard deviation of the differential count of leukocytes of Jafarabadi buffaloes and half-breed from the seventh to the 300th day of life, dewormed on the seventh day of life and subsequently every 60 days with 200mcg of ivermectin, in Ranch A, subcutaneously

Group	Age (days)	Nº	Leukocytes (X10³/mm³)	Lymphocytes	Monocytes	Segmented neutrophils	Stab neutrophils	Eosinophils	Basophils
G1	7	2	11.75±4.31	35.50±4.94	4.00±0.00	57.00±1.41	2.50±2.12	1.00±1.41	0.00
G1	15	2	10.95±0.07	57.00±18.3	3.50±4.94	37.50±21.92	1.50±2.12	0.50±0.70	0.00
G1	30	2	15.15±3.88	66.00±4.24	4.50±6.36	23.50±6.36	3.00±0.00	0.50±0.70	2.50±3.53
G1	60	4	13.50±1.33	67.25±8.95	3.25±2.50	26.75±6.65	0.00±0.00	1.50±1.29	1.25±1.89
G1	90	4	14.40±1.92	50.25±17.6	3.75±2.62	19.25±24.48	3.75±3.50	23.00±25.41	0.00
G1	120	5	12.22±1.91	60.20±8.25	8.60±8.70	26.80±10.20	2.40±1.51	1.00±1.41	1.00±1.22
G1	150	1	15.60	80	3	16	1	0	0
G1	180	4	11.14±2.74	72.25±7.04	2.00±2.82	20.75±7.04	3.75±2.98	1.25±1.50	0
G1	210	4	12.20±3.52	66.25±18.5	1.00±1.41	28.00±17.26	1.75±1.50	2.75±2.06	0.25±0.50
G1	240	6	13.88±2.52	61.00±12.74	3.83±2.04	32.00±11.91	1.16±1.94	1.83±1.47	0.16±0.40
G1	270	5	13.52±2.75	63.20±6.37	7.00±8.24	24.40±4.82	1.20±1.64	4.00±3.74	0.25±0.50
G1	300	3	15.14±2.25	71.66±13.57	3.00±3.00	16.66±8.96	5.33±3.05	2.66±3.05	0.66±0.57

Table 9: Mean values and standard deviation in differential count of leukocytes of Jafarabadi buffaloes and half-breed from the seventh to the 300th day of life dewormed on the seventh day of life and subsequently every 60 days with 200mcg of ivermectin, in Ranch B, subcutaneously

Group	Age (days)	Nº	Leukocytes (X10³/mm³)	Lymphocytes	Monocytes	Segmented neutrophils	Stab neutrophils	Eosinophils	Basophils
G4	7	1	10.20	62.00	3.00	32.00	3.00	0	0
G4	15	3	12.30±3.55	56.33±9.23	1.33±0.57	32.66±8.96	4.66±2.08	4.00±6.08	1±1
G4	30	3	10.33±1.50	62.33±10.01	2.00±2.64	33.00±13.52	0.66±0.57	1.00±0	1±1
G4	60	9	12.76±3.53	60.55±9.61	2.22±1.30	34.33±10.98	1.00±1.11	1.44±1.87	0.44±0.72
G4	90	7	20.38±17.79	62.71±14.05	1.71±1.49	32.00±13.24	1.57±1.71	1.14±1.34	0.85±1.06
G4	120	6	15.36±3.14	67.33±15.21	2.00±2.09	26.50±15.83	1.33±1.75	2.50±1.87	0.33±0.511
G4	150	8	15.62±7.54	65.87±7.86	3.37±2.50	28.00±7.42	0.75±1.16	1.87±1.45	0.12±0.35
G4	180	8	15.50±4.13	72.62±5.65	2.50±2.44	21.62±4.43	1.12±1.55	2.00±1.85	0.12±0.35
G4	210	11	14.99±4.09	69.09±12.26	4.00±2.79	23.81±7.80	1.27±2.24	1.271.42	0.54±1.21
G4	240	8	15.62±2.60	68.75±13.02	3.87±2.85	23.87±8.70	1.62±2.44	1.75±1.75	0.12±0.35
G4	270	8	16.67±4.16	65.25±14.86	3.375±3.4	27.12±11.72	2.12±2.53	1.37±1.18	0.75±0.46
G4	300	8	14.97±0.94	59.20±14.91	2.60±2.75	31.60±12.45	3.30±2.40	2.40±2.54	0.90±1.10

Table 10: Mean values and standard deviation of differential count of leukocytes of Jafarabadi buffaloes and half-breed from the seventh to the 300th day of life dewormed in Ranch A on the 15th, 30th, 60th, and 180th days of life with 10mg of fenbendazole per kg of live weight, orally

Group	Age (days)	Nº	Leukocytes (X 10³/mm³)	Lymphocytes	Monocytes	Segmented neutrophils	Stab neutrophils	Eosinophils	Basophils
G2	15	2	14.60±1.41	55.50±0.70	1.50±0.70	39.50±3.53	2.50±0.70	0±0	1.00±1.41
G2	30	4	12.10±3.98	76.75±4.27	4.25±2.06	15.75±5.12	1.50±1.91	0.75±1.50	1.00±1.41
G2	60	7	13.90±5.08	63.71±10.07	1.71±1.25	26.42±15.09	2.42±2.22	5.71±12.52	0
G2	90	7	18.11±2.80	63.57±13.42	2.42±2.87	26.57±11.35	3.42±2.43	3.57±3.10	0.42±0.78
G2	120	4	17.65±1.90	55.25±18.50	3.50±4.35	35.25±10.21	4.25±7.18	1.75±2.36	0.00
G2	150	3	13.75±2.57	70.66±1.52	0.66±1.15	24.66±2.88	1.00±1.73	2.00±1	1±1
G2	180	5	13.30±1.28	64.80±12.77	3.60±2.50	27.00±13.61	3.20±4.54	1.80±1.09	1.60±2.60
G2	210	7	14.27±2.59	65.71±17.42	3.85±3.89	27.57±17.17	1.00±1.41	1.71±1.88	0.14±0.37
G2	240	8	14.76±2.50	57.00±11.16	2.12±1.80	37.12±9.64	0.75±0.70	2.37±2.44	0.62±0.74
G2	270	3	13.20±2.74	57.00±5.29	5.66±5.68	32.66±6.02	2.33±4.04	2.33±1.15	0.00
G2	300	7	14.45±2.29	58.85±9.54	3.85±3.84	31.00±10.44	3.85±1.06	1.42±0.97	1.00±1.00

Table 11: Mean values and standard deviation of differential count of leukocytes of Jafarabadi buffaloes and half-bree from the seventh to the 300th day of life dewormed in Ranch B on the 15th, 30th, 60th and 180th day of life with 10mg of fenbendazole per kg of live weight, orally

Group	Age (days)	Nº	Leukocytes (X 10^3/mm^3)	Lymphocytes	Monocytes	Segmented neutrophils	Stab neutrophils	Eosinophils	Basophils
G5	7	2	14,97±2,48	64.50±3.53	0.50±0.70	32.00±5.65	1.50±0.70	0.50±0.70	1.00±1.41
G5	15	4	9,20±2,96	58.25±16.23	2.25±1.50	35.75±16.85	2.25±1.50	0.50±1.00	1.00±0.81
G5	30	4	11,77±2,29	61.75±13.93	3.00±2.16	32.50±13.79	1.75±2.06	1.00±0.81	0
G5	60	11	11,12±2,54	58.90±15.00	2.36±1.50	34.45±12.93	1.36±2.65	2.54±2.11	0.36±0.50
G5	90	8	15,50±3,46	57.00±11.55	1.75±1.58	37.12±10.94	2.12±2.16	1.25±1.48	0.75±1.03
G5	120	6	16,43±1,57	60.66±18.83	0.16±0.40	34.83±20.19	2.00±1.41	2.00±1.67	0.33±0.81
G5	150	7	17,58±2,22	70.28±8.73	1.57±0.53	23.71±8.22	1.85±1.57	2.28±2.42	0.28±0.48
G5	180	10	14,07±3,93	74.40±7.33	1.10±1.28	21.40±6.70	1.00±1.15	1.60±1.17	0.50±0.84
G5	210	12	14,61±1,73	64.50±8.54	3.50±2.71	27.58±7.22	2.08±2.46	1.58±1.62	0.75±0.96
G5	240	9	15,40±3,21	65.55±5.45	3.77±2.68	27.11±5.08	2.11±2.89	0.88±1.36	0.55±1.01
G5	270	10	15,77±3,47	59.30±11.12	1.70±1.70	30.70±9.49	6.10±4.93	1.20±1.98	1.00±1.24
G5	300	12	15,08±2,53	59.25±17.24	2.58±1.92	31.50±12.88	4.75±6.15	1.33±1.49	0.58±0.79

Table 12: Mean values and standard deviation of differential count of leukocytes of Jafarabadi buffaloes and half-breed from the seventh to 300th day of life wormed in Ranch A

Group	Age (days)	Nº	Leukocytes (X10^3/mm^3)	Lymphocytes (%)	Monocytes (%)	Segmented neutrophils (%)	Stab neutrophils (%)	Eosinophils (%)	Basophils (%)
G3	7	4	12.67±3.29	49.75±17.95	5.00±3.46	41.50±15.26	2.75±2.87	1.00±1.15	0
G3	15	2	15.15±3.04	34.00±4.24	0.50±0.70	56.50±4.94	7.00±7.07	2.00±2.82	0
G3	30	3	13.02±3.80	75.66±7.63	5.00±2.00	14.66±6.65	4.00±2.00	0.33±0.57	0.33±0.57
G3	60	9	16.61±4.83	56.00±8.88	2.88±3.17	32.77±13.89	1.44±1.66	6.33±11.01	0.55±1.33
G3	90	5	16.18±4.42	64.20±13.91	3.00±2.54	21.40±12.09	4.00±3.31	5.60±4.82	1.80±2.68
G3	120	5	14.45±4.82	68.20±14.14	3.20±1.92	21.00±13.92	1.60±1.81	5.40±5.54	0.60±0.54
G3	150	7	15.78±2.31	60.14±12.23	3.42±1.81	28.85±10.62	2.42±2.29	4.14±1.86	1.00±1.15
G3	180	10	14.65±2.85	65.00±8.70	2.60±2.11	27.00±9.04	1.50±0.97	3.40±2.22	0.50±0.70
G3	210	10	15.56±1.80	65.90±11.47	2.00±1.05	27.40±11.09	1.80±1.87	2.40±2.41	0.50±0.84
G3	240	9	17.28±1.79	53.11±13.21	3.00±2.00	33.33±8.66	2.88±2.47	6.22±5.51	1.44±1.81
G3	270	10	15.82±2.66	56.30±14.35	2.10±2.72	32.00±12.19	3.40±5.37	5.80±3.99	0.40±0.84
G3	300	6	15.48±3.61	58.50±12.53	4.33±4.03	28.33±11.36	4.00±3.84	4.33±1.63	0.50±0.83

Table 13: Mean values and standard deviation of differential count of leukocytes of Jafarabadi buffaloes and half-breed from the seventh to the 300th day of life, wormed in Ranch B

Group	Age (days)	Nº	Leukocytes (X10^3/mm^3)	Lymphocytes (%)	Monocytes (%)	Segmented neutrophils (%)	Stab neutrophils (%)	Eosinophils (%)	Basophils (%)
G6	7	3	15,34±2,38	54.00±9.64	2.00±1.73	40.00±13.07	2.00±1	1.33±1.15	0.66±0.57
G6	15	2	9,60±2,40	52.00±7.07	2.00±1.41	39.50±13.43	4.00±4.24	1.00±1.41	1.50±0.70
G6	30	4	11,90±3,39	62.50±14.97	3.00±2.16	32.25±16.33	1.00±1.41	1.00±1.41	0.25±0.50
G6	60	7	13,08±2,67	60.00±7.37	4.00±1.52	32.42±6.77	1.14±1.46	2.14±1.46	0.28±0.48
G6	90	4	14,14±2,80	61.00±7.11	1.25±1.25	33.25±6.29	1.75±1.70	1.50±0.57	1.25±0.95
G6	120	7	18,10±3,83	66.42±14.84	2.00±1.15	28.28±14.46	1.42±1.13	1.71±1.11	0.14±0.37
G6	150	6	13,25±3,92	66.00±17.57	10.00±20.65	20.16±14.78	1.33±1.96	2.33±1.50	0.16±0.40
G6	180	10	14,03±2,76	67.90±12.49	5.10±3.57	23.50±9.05	1.70±2.45	1.30±1.49	0.50±0.52
G6	210	7	14,10±1,70	73.57±10.69	2.71±2.62	20.42±8.97	1.14±1.06	1.85±1.57	0.28±0.48
G6	240	7	14,60±3,51	64.85±8.21	4.00±2.38	25.14±5.84	2.85±3.93	2.71±2.05	0.42±0.78
G6	270	11	17,45±3,31	59.18±21.31	8.27±18.13	27.72±8.12	3.27±2.00	1.54±1.57	0
G6	300	9	14,93±2,47	59.11±8.20	2.00±1.73	34.55±7.12	2.22±2.38	1.33±1.50	0

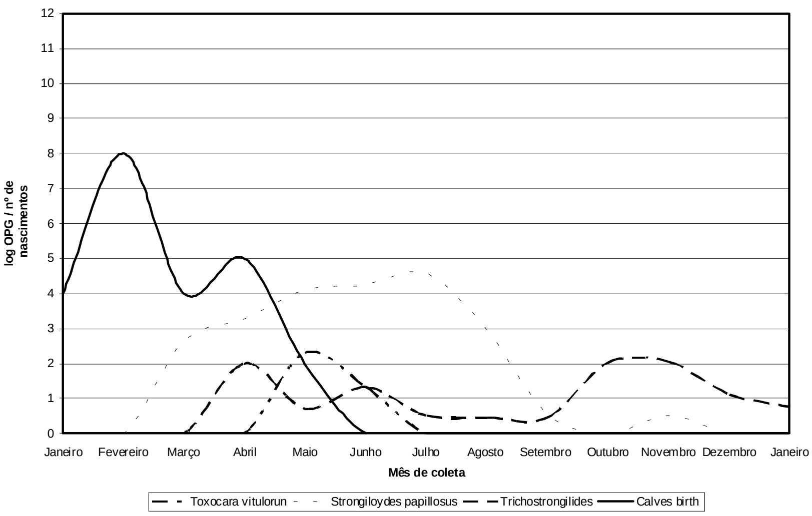

Figure 1: Birth number and nematodes EPG - with logarithm transformation of buffaloes belonging to control group from the seventh to the 300th day of age in January, 2003 and January, 2004

Haemonchus placei in bovines. Ovines highly wormed by *Haemonchus contortus* develop acute hypochromic normocytic anemia with evident increase of erythropoiesis. Animals chronically infected by *Haemonchus* spp. present normochromic normocytic or hypochromic microcytic anemia, with little or no presence of reticulocytes (Jain, 1986).

The higher number of EPG observed in Group 1, 2, and 3 animals in relation to Groups 4, 5, and 6 animals is probably the outcome of the raising system, to wit, a lower volume of milk intake by Ranch A animals, causing the reduction of the natural capacity of developing resistance to the diagnosed helminthes (Table 1).

Associated to resistance decrease, the competition for nutrients and further spoliative action mechanisms of parasites on hosts are responsible for the significant decreases in the erythrocytes count, in the hematocrit, and in MCHC ($P<0.01$), in hemoglobin, and in MCV ($P<0.05$).

A significant difference ($P<0.01$) in relation to MCV in animals of different genders was also observed.

The EPG increase of G2 after 180th day of life in Ranch A presented in feces and the decreased number of erythrocytes in the circulating blood were possibly due to the higher sensitivity of G2 animals to reinfestation by ingested helminthes larvae as compared to the G1, G3, G4, G5, and G6 animals. This generally occurs due to the absence of larvae stimulation able to trigger the development of immune response during the first months of life (Guimarães, et al. 1975, Levine, 1980, Lloyde and Solsby, 1987).

The weather conditions observed during the period – accumulated rainfall of 120mm and mean temperature of 24°C – allowed the larval development, eclosion, and transfer period of nematode parasites in the gastrointestinal treat, infective from within the eggs present in feces. The control animals also presented an average concentration increase of EPG in October, which reflects the parasite challenge by trichostrongylus helminth parasites to which the animals were submitted in September.

Buffaloes from the Brazilian Southeast present reproductive seasonality and a consequent concentration of birth rate in February and March (Baruselli, 2005) (Fig.1). This fact precludes the use of the strategic treatment for the control of all the endoparasites identified in this study as proposed by Láu (1999), foreseeing medication interruption on the 180th day of life, an age coinciding with the onset of spring in the Central and Southeastern Brazilian regions.

In spite of the previous reduction, the erythrogram values in wormed animals remained within the normal limits for the buffalo species according to Láu (1985) and Jain (1986) throughout the experimental stage. Probably, the reduced period of exposition to the infection *versus* the evaluation and the low parasitic level were not enough for the animals to develop acute or chronic anemia.

The results found in these animals do not exclude the possibility of young buffaloes to develop anemia due to parasitism by the regional nematode species. The mean values of the hematological indexes found in all animals are presented (Tables 2 to 7) according to the groups they belonged.

In buffaloes, as occur in bovines (Hammenberg, 1986), the presence of parasite in the gastrointestinal tract is responsible for the competition for food, anorexia, digestion capacity decrease, limited protein and iron absorption, and for the blood spoliation of the wormed animals.

Discrete variations of the number of leukocytes and the proportion of different cell types along to the development of the animals are a normal response regarding all the species (Jain, 1986). The results found by the present work, to the exception of eosinophils, agree with the mean values for buffaloes of the same age reported by Santos (1984), Jain (1986) and Bonfim (1995).

The presence of parasites in the gastrointestinal treat of buffaloes is responsible for the hematological parameters changes, which can reduce the capacity of animal development and production. The intensity of these changes and the type of immune response of animals are proportional not only to the level of parasitic challenge with the animals are exposed to, but also to their age, parasite species present in the property, time of the year, and techniques of nutritional management, besides the parasitic control adopted by the production system.

Once the normal hematological parameters and related alterations changes of the wormed buffaloes are known, the veterinarian is able to dispose of his diagnosis hypothesis other diseases presenting clinical evolution similar to parasitoses.

Conclusion

This study confirmed the interference of gastrointestinal nematodes present in the intestines of buffaloes younger than a year in blood profile. Wormed animals presented reduced serum concentration of hemoglobin, hematocrit, total serum protein concentration, and leukocytosis with eosinophilia. The variation intensity of the studied parameters varied according to the number and species of parasites present in the gastrointestinal treat of calves in the different ages.

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