Medicina Veterinária (UFRPE) ISSN 2675-6617

Blood and biochemical parameters in young horses raised in the semiarid region along the year

[*Parâmetros hematológicos e bioquímicos de cavalos jovens criados na região semiárida ao longo do ano*]

"Scientific Article/Artigo Científico"

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Abstract
Seasonality is a characteristic that influences forage availability and quality. Research has been conducted to correlate seasonal variations with the modification of metabolic parameters in horses. Despite the adaptive capacity of animals, extreme conditions affect animal health and well-being. Knowledge of such information contributes to greater creative efficiency. This study aimed to determine the concentration of hematological, biochemical, and mineral biomarkers in fillies over a period of 12 months. From a batch of 40 animals, 25 clinically healthy *Mangalarga Marchador* fillies were used. Colts were performed once a month for 12 months, with the animals fasted overnight but with free access to water. The results were analyzed using the SigmaPlot 13.0 program for Windows® (Systat Software, Inc.), using analysis of variance with two factors and Tukey's test. The significance level was set at 5%. Results are expressed as means ± standard error of the mean. Although no clinical changes were observed in the animals, the findings of this study may reflect their general health status. Several species, such as horses, are subject to variation in physiological processes without denoting pathological states. From the data obtained, it can be concluded that the animals in this study were clinically healthy, and these data can be used as parameters for the evaluation of animals of the *Mangalarga Marchador* breed.

Keywords: forage seasonality; metabolic profile; yearling; welfare.

Resumo
A sazonalidade é uma característica que influencia a disponibilidade e qualidade da forragem. Pesquisas têm sido conduzidas para correlacionar variações sazonais com a modificação de parâmetros metabólicos em cavalos. Apesar da capacidade adaptativa dos animais, condições extremas afetam a saúde e o bem-estar animal. O conhecimento dessas informações contribui para uma maior eficiência criativa. Este estudo teve como objetivo determinar a concentração de biomarcadores hematológicos, bioquímicos e minerais em potras durante um período de 12 meses. De um lote de 40 animais, foram utilizadas 25 potras *Mangalarga Marchador* clinicamente saudáveis. As coletas foram realizadas uma vez por mês durante 12 meses, com os animais em jejum durante a noite, mas com livre acesso à água. Os resultados foram analisados por meio do programa SigmaPlot 13.0 para Windows® (Systat Software, Inc.), por meio de análise de variância com dois fatores e teste de Tukey. O nível de significância foi estabelecido em 5%. Os resultados são expressos como média ± erro padrão da média. Embora não tenham sido observadas alterações clínicas nos animais, os achados deste estudo podem refletir seu estado geral de saúde. Diversas espécies, como os cavalos, estão sujeitas a variações nos processos fisiológicos sem denotar estados patológicos. A partir dos dados obtidos, pode-se concluir que os animais deste estudo estavam clinicamente saudáveis, e esses dados podem ser utilizados como parâmetros para avaliação de animais da raça *Mangalarga Marchador*.

Palavras-chave: sazonalidade da forragem; perfil metabólico; sobreano; bem-estar.

Received 29 May 2022. Accepted 14 August 2022.
DOI: https://doi.org/10.26605/medvet-v16n2-4999

Introduction

Currently, there is a search for breeding systems in which horses receive more forage to improve general health, keeping animals with more natural nutrition and favoring a balanced microbiota (Brandi and Furtado, 2009; Grimm et al., 2017; Raspa et al., 2022). However, seasonal forage availability can influence both the microcrobiota (Salem et al., 2018**)** and thus variations in the concentrations of metabolic biomarkers in young horses that live in different breeding systems and thus interfere with athletic health, well-being and longevity.

Research relating to possible variations in metabolic biomarkers with annual forage availability and seasonality for horses is scarce, primarily in animals kept on pasture systems and with forage-based feeding programs. Horses have an important adaptive capacity to these systems, where the energy and protein bases are forage are major nutrients' source (Salem et al., 2018; Raspa et al., 2022); however, during extreme environmental conditions the pasture quality decreased so much and have relationship with reduction of horses' health and well-being. Long-term studies to assess these metabolic parameters are not common; however, some studies follow horses for periods longer than 3 or 5 months to verify the effects of forage and food seasonality on metabolic biomarkers (Piccione et al., 2001; Ali et al., 2004; Piccione et al., 2004; Piccione et al., 2005; Satué et al., 2013).

To test the hypothesis that different variables such as environment, management, and nutrition modify young athletic horses' biomarkers throughout the seasons and months of the year, in the semiarid region of Pernambuco and in semi-extensive breeding, a study was conducted that aimed to determine the concentrations of hematological biochemicals, and minerals biomarkers, over 12 months in young fillies *Mangalarga Marchador*. Knowing this information can contribute to the future athletic performance, reduction of costs, and welfare of the herd.

Material and Methods

Animals and management practices

From a batch of 40 field animals, 25 fillies of the *Mangalarga Marchador* breed (>24 months and <42 months; 350 kg) were used, which were followed between 10 and 12 months in the same year. They were clinically healthy, under the same management, including vaccinations and deworms applications, and nutrition programs. They were housed in Gravatá-PE (Lat: -8.2096, Long: -35.5695; mean annual precipitation: 800 mm). They were supplemented with concentrate produced on the farm (2.0 kg/animal/day; dry matter: 89.26%; ether extract: 1.66%; crude protein: 8.42%; gross energy: 3.79 Mcal/kg; mineral matter: 2.08%; neutral detergent fiber: 32.71%; acid detergent fiber: 22.98%) and had free access to Buffel grass pasture (*Cenchrus ciliaris* cv. Biloela) in the rainy season (April-September) and supplementation with whole corn silage (3.0–4.0 kg/day/animal) plus farm's concentrate in the dry period (October-March). Mineralized salt (Co-Equi Tortuga/DSM), and water were provided ad libitum. Blood samples were collected after a day of rest, if they do any type of exercise.

Collection and laboratory analysis

They were performed once a month for 12 months, with the animals fasted overnight but with free access to water. Body mass was measured on an electronic scale, and blood samples were obtained in vacuum tubes containing EDTA for hematological examination using semi-automatic equipment (Roche® Poch 100iv) (red blood cells, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin concentration, erythrocyte distribution amplitude standard deviation, erythrocyte distribution amplitude coefficient of variation, leukocytes, lymphocytes, platelets, and other cells). For biochemical analysis, samples were collected in tubes without anticoagulant to obtain: serum protein, albumin, triglycerides, high-density lipoprotein, iron, calcium, phosphorus, magnesium, and chloride; the analyses were performed in semi-automatic equipment (Doles® D-250), using commercial kits and following the manufacturer's recommendations.

Statistical analysis

The results were analyzed using the SigmaPlot 13.0 program for Windows® (Systat Software, Inc.), using analysis of variance with two factors (season and months of the year) and Tukey's test. In both cases, the significance level was set to 5%. All results are expressed as means ± standard error.

Results

The results the ANOVA with 2 ways showed that body mass and hematological parameters did not show significant variation for seasons, and interactions (p>0.05), except to chlorides and triglycerides (Table 1). Chloride concentration varied according to the season and month with significant interaction (P<0.05) (Table 1) and this mineral concentration were higher in the summer and autumn then in the winter and spring seasons (Table 2). Triglycerides varied by month and showed significant interaction (P<0.05) (Table 1). Also, chloride concentrations from January to June were also lower than those observed from July to December (P<0.05) (Table 3). Triglycerides concentrations were lower at the beginning of the year, rising in the middle, and decreasing at the end of the year (P<0.05) (Table 3). The results that body mass and hematological parameters did not show significant variation for month in the year (p>0.05) (Table 4).

Table 1. Results of Two way-way analysis of variance with 2 factors and the interaction of seasons versus months of each dependent variable on body mass, hematological and blood biochemistry profile, during a whole year period of fillies between 2-3 years of age in the semiarid region.

Variable	Seasons	Months	Interaction seasons x months
Body mass (kg)	*P* >0,05	*P* >0,05	*P* >0,05
Erythrocyte (x10^6/µL)	*P* >0,05	*P* >0,05	*P* >0,05
Hemoglobin (g/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Hematocrit (%)	*P* >0,05	*P* >0,05	*P* >0,05
VCM (fL)	*P* >0,05	*P* >0,05	*P* >0,05
CHCM (g/dl)	*P* >0,05	*P* >0,05	*P* >0,05
RDW-SD (fL)	*P* >0,05	*P* >0,05	*P* >0,05
RDW-CV (%)	*P* >0,05	*P* >0,05	*P* >0,05
Leukocytes (x10^3/µL)	*P* >0,05	*P* >0,05	*P* >0,05
Lymphocytes (x10^3/µL)	*P* >0,05	*P* >0,05	*P* >0,05
Platelets (x10^3/µL)	*P* >0,05	*P* >0,05	*P* >0,05
Other cells (x10^3/µL)	*P* >0,05	*P* >0,05	*P* >0,05
Serum protein (g/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Albumin (g/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Triglycerides (mg/dl)	*P* >0,05	*P* <0,05	*P* <0,05
Cholesterol (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
HDL (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Iron (µg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Calcium (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Ionized calcium (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Phosphorus (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Magnesium (mg/dl)	*P* >0,05	*P* >0,05	*P* >0,05
Chloride (mg/dl)	*P* <0,05	*P* <0,05	*P* <0,05

Observations: VCM: mean corpuscular volume; CHCM: mean corpuscular hemoglobin concentration; RDW-SD: red blood cell distribution index – standard deviation; RDW-CV: distribution index and red blood cells – coefficient of variation; Other cells: neutrophil and monocyte; HDL: high density cholesterol.

Table 2. Results of two-way analysis of variance for different seasons on the body mass, hematological and blood biochemistry biomarkers profile, during a whole year period of fillies between 2-3 years of age in the semiarid region.

Parameters	Seasons			
	Summer	Autumn	Winter	Spring
Body mass (kg)	345,90±24,05	353,56±16,15	334,31±16,87	315,31±25,80
Erytrocyt (x10^6/µL)	8,01±1,15	8,34±0,77	8,34±0,811	8,59±1,24
Hemoglobin (g/dl)	11,20±1,53	11,36±1,02	11,61±1,07	12,61±1,64
Hematocrit (%)	36,40±4,75	36,24±3,19	33,99±3,33	36,24±5,10
VCM (fL)	41,67±1,82	40,17±1,22	38,92±1,28	40,99±1,96
CHCM (g/dl)	31,61±0,82	32,45±0,55	33,70±0,57	34,45±0,88
RDW-SD (fL)	32,45±2,08	33,62±1,39	36,12±1,46	35,62±2,23
RDW-CV (%)	20,92±1,39	21,75±0,93	21,75±0,97	20,00±1,49
Leukocytes (x10^3/µL)	12,51±3,02	13,67±2,03	8,92±2,12	10,17±3,24
Lynfocytes (x10^3/µL)	6,79±2,16	6,96±1,45	5,21±1,52	5,96±2,32
Platelets (x10^3/µL)	152,29±48,07	151,96±32,28	91,46±33,72	183,71±51,58
Other cells (x10^3/µL)	5,82±1,29	6,49±0,86	2,99±0,90	2,74±1,38
Serum protein (g/dl)	10,65±1,27	8,82±0,85	11,32±0,89	12,57±1,36
Albumin (g/dl)	3,24±0,50	2,90±0,33	2,40±0,35	2,40±0,54
Triglycerides (mg/dl)	59,57±10,83	48,07±7,27	38,32±7,59	34,32±11,62
Cholesterol (mg/dl)	93,79±15,50	86,79±10,41	117,54±10,87	118,54±16,63
HDL (mg/dl)	62,58±13,41	46,58±9,00	73,58±9,04	57,33±14,39
Iron (µg/dl)	136,57±22,99	136,74±15,44	129,99±16,13	95,74±24,67
Calcium (mg/dl)	9,43±1,13	10,77±0,75	11,21±0,79	11,43±1,21
Ionized calcium (mg/dl)	4,74±0,88	5,64±0,59	5,99±0,62	5,91±0,95
Phosphorus (mg/dl)	5,36±0,89	4,52±0,59	4,77±0,62	5,77±0,95
Magnesium (mg/dl)	7,79±0,68	8,46±0,46	7,46±0,48	7,96±0,73
Chloride (mg/dl)	144,42±3,17 AB	146,42±2,13 A	123,17±2,22 B	124,67±3,40 B

Observations: Different letters on the same line indicate $p<0.05$. VCM: mean corpuscular volume; CHCM: mean corpuscular hemoglobin concentration; RDW-SD: red blood cell distribution index – standard deviation; RDW-CV: distribution index and red blood cells – coefficient of variation; Other cells: neutrophil and monocyte; HDL: high density cholesterol.

Discussion

Throughout this experiment, the parameters evaluated did not vary significantly, which was unexpected due to the climatic and nutritional characteristics of the semiarid region, probably because these young fillies had well-balanced nutritional and management programs. Using strategic supplementation with silage and concentrate in the dry and rainy seasons may have contributed to the positive results in the development and health of the animals.

The four seasons are not well defined in the semiarid regions; however, there is a rainy period (increased flowering, fruit, and forage availability), a dry period (dry forage and loss of foliage), and transition periods between them. At the site of this experiment, summer and spring were dry, with loss of foliage associated with a short period of dry fruit drops, mostly mesquite (*Prosopois juliflora*), whereas in autumn and winter, there was an increase in flowering, native forage, and rainfall.

Table 3. Results of least square mean by two-way analysis of variance for different months of the year on the blood biochemistry biomarkers and mineral profile, during a whole year period of fillies between 2-3 years of age in the semiarid region.

Variable	Months of the Year											
	1	2	3	4	5	6	7	8	9	10	11	12
Serum protein (g/dl)	10,04 ±1,37	10,68 ±1,39	10,90 ±1,37	13,18 ±0,93	12,22 ±1,03	11,02 ±1,12	10,02 ±0,86	10,92 ±1,03	11,32 ±1,03	9,52 ±1,24	9,47 ±1,43	10,77 ±1,41
Albumin (g/dl)	1,78 ±0,54	2,00 ±0,55	2,07 ±0,54	2,50 ±0,37	2,43 ±0,40	2,16 ±0,44	2,83 ±0,34	3,33 ±0,40	3,93 ±0,40	3,33 ±0,49	3,33 ±0,57	3,16 ±0,56
Tryglicerides (mg/dl)	16,07 ±11,74 BC	26,83 ±11,86 BC	28,07 ±11,74 BC	32,50 ±7,96 BC	31,40 ±8,77 BC	44,00 ±9,61 C	48,00 ±7,33 C	85,75 ±8,77 AC	59,15 ±8,77 C	57,75 ±10,57 C	58,15 ±12,22 C	53,25 ±12,06 C
Cholesterol (mg/dl)	96,94 ±16,81	110,70 ±16,98	112,37 ±16,81	118,37 ±11,40	120,97 ±12,55	133,04 ±13,76	101,37 ±10,49	103,42 ±12,55	91,42 ±12,55	90,62 ±15,13	86,82 ±17,49	83,95 ±17,27
HDL (mg/dl)	37,58 ±14,54	33,77 ±14,69	43,29 ±14,54	54,43 ±9,86	56,03 ±10,86	57,77 ±11,90	38,43 ±9,08	67,43 ±10,86	73,03 ±10,86	86,43 ±13,09	89,68 ±15,13	82,35 ±14,94
Iron (µg/dl)	108,61 ±24,94	141,02 ±25,18	120,75 ±24,94	105,18 ±16,91	94,42 ±18,62	106,35 ±20,41	111,02 ±15,56	134,97 ±18,62	120,37 ±18,62	125,77 ±22,44	142,02 ±25,95	186,68 ±25,61
Calcium (mg/dl)	10,58 ±1,22	10,41 ±1,23	11,55 1,27	11,90 ±0,83	12,71 ±0,91	10,44 ±1,00	10,90 ±0,76	10,33 ±0,91	10,42 ±0,91	10,00 ±1,10	9,53 ±1,27	10,54 ±1,26
Ionized calcium (mg/dl)	5,93 ±0,96	6,56 ±0,97	5,99 ±0,96	5,71 ±0,65	6,68 ±0,71	5,66 ±0,78	5,68 ±0,59	4,97 ±0,71	4,72 ±0,71	4,92 ±0,86	4,73 ±0,99	5,31 ±0,98
Phosphorus (mg/dl)	4,17 ±0,96	4,41 ±0,97	5,32 ±0,96	5,75 ±0,65	6,18 ±0,72	4,91 ±0,79	5,58 ±0,60	5,93 ±0,72	5,73 ±0,72	4,33 ±0,86	4,33 ±1,00	4,66 ±0,99
Magnesium (mg/dl)	7,69 ±0,74	8,29 ±0,75	7,83 ±0,74	8,12 ±0,50	8,25 ±0,55	7,79 ±0,61	8,45 ±0,46	8,05 ±0,55	8,65 ±0,55	7,45 ±0,67	7,15 ±0,77	7,29 ±0,76
Chloride (mg/dl)	128,96 ±3,44 BC	125,91 ±3,47 B	124,39 ±3,44 B	124,25 ±2,33 BD	126,45 ±2,57 B	125,91 ±2,81 B	147,25 ±2,14 A	144,90 ±2,57 A	144,70 ±2,57 A	143,50 ±3,09 AC	140,40 ±3,58 ABC	139,50 ±3,53 ABC

Observations: Different letters on the same line indicate p<0.05. HDL: high density cholesterol.

Body mass, hematological, and biochemical profiles are important to assess the general health of animals, and are influenced by the environment, management, and nutrition. Monthly and seasonal variations as well as fluctuations in temperature, humidity, and precipitation throughout the day can alter these parameters (Greppi et al., 1986). Apparently, the current experimental animals had good adaptive capacity to the environment and rearing system where they lived. Although not clinically significant, the findings of this study may reflect the general health of the animals, as well as the physiological behavior of their metabolism throughout our evaluation under semiarid conditions.

The results of studies in other countries may not be applicable to local conditions due to the influence of factors such as environment and management (Gul et al., 2007), suggesting the need for further research in different regions and their characteristic ecosystems to demonstrate how horses balance their blood and biochemical parameters throughout the year. The reference intervals described in most of the literature on hematological and biochemical profiles of horses refer to adult animals, as described by Stockham (1995). This pattern can still be observed today when relating different breeds, such as *Brasileiro de hipismo*, Standardbred, and *Mangalarga Marchador* horses, described by Santiago et al. (2013), Mariella et al. (2014), and Ferreira et al. (2017), respectively. Thus, age is another important variable that should be considered when evaluating hematological and biochemical parameters in horses (Gul et al., 2007). Considering these and other blood profiles can help in the development of therapeutic and nutritional protocols, as well as in evaluating the physiological and pathological states of animals in different age groups.

Table 4. Results of two-way analysis of variance for different months of the year on the body mass and hematological biomarkers profile, during a whole year period of fillies between 2-3 years of age in the semiarid region.

Parameter	Months of the Year											
	1	2	3	4	5	6	7	8	9	10	11	12
Body mass (kg)	328,52 ±26,08	319,04 ±26,34	335,94 ±26,08	329,37 ±17,68	335,10 ±19,48	337,04 ±21,34	327,70 ±16,28	350,55 ±19,48	359,35 ±19,48	346,95 ±23,47	338,75 ±27,14	338,95 ±26,79
Erytrocyt ($x10^6/\mu L$)	8,17 ±1,25	8,97 ±1,26	9,31 ±1,25	8,31 ±0,85	7,77 ±0,78	8,31 ±1,02	7,97 ±1,12	8,37 ±0,93	8,57 ±0,93	7,97 ±1,12	7,72 ±1,30	8,39 ±1,28
Hemoglobin (g/dl)	11,50 ±1,66	13,16 ±1,67	13,07 ±1,66	12,50 ±1,12	11,13 ±1,24	11,66 ±1,35	11,33 ±1,03	11,68 ±1,24	11,88 ±1,24	11,08 ±1,49	10,48 ±1,72	10,91 ±1,70
Hematocrit (%)	34,31 ±5,15	38,31 ±5,20	39,02 ±5,15	36,31 ±3,49	33,47 ±3,85	35,14 ±4,22	35,47 ±3,22	36,12 ±3,85	37,12 ±3,85	34,72 ±4,64	33,27 ±5,36	35,31 ±5,29
VCM (fL)	40,75 ±1,98	40,91 ±2,00	40,03 ±1,98	40,75 ±1,34	40,85 ±1,48	40,25 ±1,62	41,25 ±1,23	40,70 ±1,48	40,90 ±1,48	39,50 ±1,78	39,50 ±2,06	39,66 ±2,03
CHCM (g/dl)	33,58 ±0,89	33,92 ±0,90	33,43 ±0,89	33,43 ±0,60	32,80 ±0,66	32,93 ±0,73	32,60 ±0,55	33,55 ±0,66	32,95 ±0,66	32,35 ±0,80	32,80 ±0,93	32,27 ±0,91
RDW-SD (fL)	38,28 ±2,25	37,50 ±2,28	35,85 ±2,25	36,00 ±1,53	34,83 ±1,41	35,16 ±1,84	34,83 ±1,41	31,53 ±1,68	31,93 ±1,68	32,33 ±2,03	32,83 ±2,35	32,33 ±2,32
RDW-CV (%)	21,61 ±1,50	20,68 ±1,52	20,61 ±1,50	20,18 ±1,02	20,35 ±1,12	20,68 ±1,23	20,35 ±0,94	20,95 ±1,12	20,75 ±1,12	22,35 ±1,35	22,50 ±1,57	22,21 ±1,55
Leukocytes ($x10^3/\mu L$)	10,67 ±3,28	12,47 ±3,31	10,95 ±3,28	9,81 ±2,22	10,04 ±2,44	11,31 ±2,68	12,64 ±2,04	11,79 ±2,44	11,39 ±2,44	11,39 ±2,95	11,74 ±3,41	11,64 ±3,36
Lymphocytes ($x10^3/\mu L$)	6,438 ±2,35	6,604 ±2,37	5,866 ±2,35	5,438 ±1,59	5,471 ±1,75	8,271 ±1,92	6,271 ±1,46	6,421 ±1,75	6,021 ±1,75	6,021 ±2,11	6,071 ±2,44	5,937 ±2,41
Platelets ($x10^3/\mu L$)	151,27 ±52,14	173,56 ±52,65	118,99 ±52,14	123,56 ±35,35	95,49 ±38,93	90,56 ±42,67	125,89 ±32,54	206,39 ±38,93	207,79 ±38,93	142,39 ±46,92	163,54 ±54,26	138,81 ±53,55
Other cells ($x10^3/\mu L$)	2,97 ±1,40	4,52 ±1,41	3,68 ±1,40	3,68 ±0,95	3,42 ±1,04	2,35 ±1,14	6,02 ±0,87	5,12 ±1,04	5,12 ±1,04	5,52 ±1,26	5,77 ±1,46	5,93 ±1,44

Observations: VCM: mean corpuscular volume; CHCM: mean corpuscular hemoglobin concentration; RDW-SD: red blood cell distribution index – standard deviation; RDW-CV: distribution index and red blood cells – coefficient of variation; Other cells: neutrophil and monocyte.

In this study, all data for hematology were within the range observed in previous studies (Holanda et al., 2013; Manso et al., 2015; Ferreira et al., 2017), which evaluated the hematological parameters of the *Mangalarga Marchador* breed. The equivalence may be associated with the similar environment and management between different studies; they also corroborate the reference data for the species (Meyer et al., 1995) which present parameters obtained under different characteristics.

Paladino et al. (2014) evaluated Standardbred horses between 3 and 10 years of age housed in Italy, and observed no difference between the values for the red series when comparing adult animals to those aged 3 years. The data observed by the author are slightly elevated when compared to those observed in this study, possibly because it was developed under very different environmental, nutritional, and management conditions, in addition to evaluating a different breed. Even subtle margins are important when evaluating the clinical picture.

Domestic animals show clear variation between species in lipoprotein profiles, and there is a significant difference in the equine species between breeds (Asadi et al., 2006). The reproductive cycle can also influence these variables, as in the present study. Protein indices reflect nutritional levels; a different diet can result in different serum protein and ALB levels. The total protein values observed were higher than those found by Manso et al. (2015) and Ferreira et al. (2017). In contrast, ALB was lower than the data from the same authors, demonstrating dietary influence, which may affect other parameters such as the lipid profile. Thus, it is possible to assess not only the physiological or pathological status but also the impact of nutritional management and physical activity programs from the biochemical profile. The metabolic profile of equines changes according to the production or consumption of these important energy metabolites.

The triglyceride levels were similar to those observed by Ferreira et al. (2017) in animals of the same breed between January and May, increasing progressively from June, with a peak observed in August and remaining at high levels until December. According to Dunkel and McKenzie (2003), triglyceride levels above the reference levels are not always associated with clinical disease. Several species, such as horses, are subject to variations in physiological processes without denoting pathological states, and long-term evaluation of these blood parameters is important to understand how horses adjust to their environment.

In contrast, when analyzing the study by Meliani et al. (2011), who evaluated triglycerides at different stages of the reproductive cycle of Arabian mares, it was observed that yearling fillies had levels closer to the lower threshold than those of other stages of the cycle. As it was developed in Algeria, changes in soil and climate, management, and nutrition have influenced the different characteristics observed. Even the lower levels found by Meliani et al. (2011) do not necessarily denote a pathological process, but rather represent organic variation in the studied group, as serum triglyceride levels reflect food intake, liver synthesis, and the animal's energy demand. As demonstrated by Barros et al. (2019), TGs are essential compounds for several metabolic mechanisms, such as the production of reproductive hormones.

When observing the mineral profile, it is commonly considered separate from the biochemical profile; however, a series of biological substances involved in the body's regulatory processes contain mineral elements such as calcium, chloride, phosphorus, and magnesium (Silva et al., 2019). The data obtained for chloride levels were moderately above the normal values described in the literature (Silva et al., 2019). Chloride is a macromineral involved in various organic functions, and its metabolism is directly linked to the activity of other mineral elements such as calcium, phosphorus, and magnesium. According to Kaneko et al. (1997), the mineral content is directly influenced by ambient temperature, considering that it is an important component eliminated during sweating in horses. Even though the study was performed in a period without radical climatic changes, climate can still significantly influence the organic metabolism of the element.

When correlated with data from Lumsdem et al. (1980), Kaneko et al. (1997), and Corley and Marr (1998), the higher chloride levels obtained are not necessarily pathological because the parameters were obtained under different environmental, management, and nutritional criteria from those mentioned in the literature. Additionally, data collection in this experiment always occurred in the morning, with the animals fasting and without intense exposure to the sun, which may reflect the levels of urinary loss and sweating, contributing to the increase in the concentration observed for the mineral in question.

Body mass is an indicator of growth; each race has its own characteristics of body development. Monitoring the weight of animals at different ages, particularly in the final period of growth, helps to assess the quality of nutritional management, which is a key feature for good performance. From the data obtained in this study, it can be observed that between 2 and 3 years of age, these females reached approximately 82% of the weight of the adult animal, considering an average of 425 kg (Cabral et al., 2004). These findings are reinforced by the results of Rezende et al. (1986), who noted that at 12 months, females of this breed reached approximately 71% of their adult weight.

Blood biochemistry assessments cannot be used as a reference in their entirety when compared to results from other countries because environmental and management changes modulate this information (Mori et al., 2003). Emphasizing the need for national research in different races, management, and environmental conditions, as well as biochemical and other physiological parameters should be encouraged.

In conclusion, no differences were observed between body mass and hematology values between the months of the year and in different seasons. Although some parameters exceeded the ranges described in the literature, they do not necessarily indicate pathological conditions and instead may be correlated with the influence of different variables. Thus, it can be inferred that the animals in this study were clinically healthy, and that these data can be used as parameters for the evaluation of animals of the *Mangalarga Marchador* breed.

Conflict of Interest

The authors declare that there is no conflict of interest.

Ethics Committee

All protocols performed were submitted to CEUA/UFRPE, to be performed under license number 085/2019.

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