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FAMACHA©: Predictive value for control of *Haemonchus* sp. in sheep from Brazilian Cerrado

FAMACHA©: Valor preditivo para o controle de *Haemonchus* sp. em ovinos do Cerrado brasileiro

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Highlights

Famacha© presented high sensitivity and specificity in sheep from Brazilian Cerrado.
Famacha© score 3 had greater sensitivity on more susceptible production categories.
Sensitivity and specificity values were similar among the three main breeds studied.

Abstract

This study aimed to evaluate the sensitivity and specificity of FAMACHA© method, correlating with packed cell volume (PCV) and egg count (FEC), as well as to evaluate the clinical signs of *Haemonchus* sp. infection in sheep from Brazilian Cerrado. Over two years (2017 to 2019), 1,435 sheep were subjected to clinical and parasitological evaluations. Sheep from six breeds (Santa Inês, Dorper, White Dorper, Ile de France, Suffolk, and crossbreed) were subdivided into five production categories (pregnant, lactating, non-pregnant/lactating ewes, breeding males, and weaned lambs). Parasitological evaluations included FEC and coproculture. In the clinical evaluation, all sheep underwent determination of the FAMACHA© score and PCV. *Haemonchus* sp. larvae were predominant in coprocultures of the flocks (76.4%) and in each animal production category evaluated (69.4 to 84.3%). FAMACHA© method showed high sensitivity (70.6%) for evaluating sheep with scores ≥ 3, and PCV < 23%, and high specificity (97.5%) in animals with higher scores (4 and 5), and PCV < 18%. A negative correlation was observed between FAMACHA© scores and PCV (-0.46) and between PCV and FEC (-0.47), while a positive correlation was observed between FAMACHA© scores and FEC (0.22) ($p < 0.01$). The vast majority of the animals evaluated (54.5%) were clinically resistant to gastrointestinal parasites. Due to the high sensitivity and specificity, we concluded that the method could be a valuable diagnostic alternative and an ancillary tool in the implementation of selective treatment for

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helminthic infection in sheep from Brazilian Cerrado.
Key words: *Haemonchus.* Selective control. Sensitivity. Small ruminants. Specificity.

Resumo

Este estudo objetivou avaliar a sensibilidade e especificidade do método FAMACHA©, e sua correlação com volume globular (VG) e contagem de ovos de nematódeos por grama de fezes (OPG), no diagnóstico de infecção parasitária por Haemonchus sp., além de avaliar os sinais clínicos frente às infecções por tais endoparasitos, em ovinos do Cerrado brasileiro. Ao longo de dois anos (2017 a 2019), o total de 1.435 ovinos foram submetidos a avaliação clínica e parasitológica. Os ovinos de seis raças (Santa Inês, Dorper, White Dorper, Ile de France, Suffolk e mestiços) foram subdivididos em cinco categorias de produção (ovelhas gestantes, lactantes, não gestantes/lactantes, machos reprodutores e cordeiros desmamados). As avaliações parasitológicas incluíram OPG e coprocultura. Na avaliação clínica, os ovinos passaram por determinação do escore FAMACHA© e do VG. As larvas de Haemonchus sp. foram predominantes nas coproculturas dos rebanhos (76,4%) e em cada categoria de produção animal avaliada (69,4 a 84,3%). O método FAMACHA© apresentou alta sensibilidade (70,6%) para avaliar animais com escores ≥ 3 e VG < 23% e alta especificidade (97,5%) em animais com escores mais elevados (4 e 5), e VG < 18%. Observou-se correlação negativa entre os escores FAMACHA© e VG (-0,46) e entre VG e OPG (-0,47), enquanto uma correlação positiva foi observada entre os escores FAMACHA© e OPG (0,22) ($p < 0.01$). A grande maioria dos animais avaliados (54,5%) mostrou-se clinicamente resistente às parasitoses gastrointestinais. Devido à alta sensibilidade e especificidade, concluímos que o método pode ser uma alternativa diagnóstica valiosa e ferramenta auxiliar na implementação do tratamento seletivo para infecção por helmintos em ovinos do Cerrado brasileiro.
Palavras-chave: Controle seletivo. Especificidade. *Haemonchus.* Pequenos ruminantes. Sensibilidade.

Introduction

Gastrointestinal parasites pose significant sanitary and financial barriers to sheep productivity (Costa & Amarante, 2015; Igarashi et al., 2013). Economic losses due to parasitism are mainly due to the limited methods of controlling infection, which focus exclusively on pharmacotherapy. The indiscriminate spending on antiparasitics drugs and their inability to control gastrointestinal parasites lead to damages that can substantially affect production (Bricarello, 2015).

The Faffa Malan Chart (FAMACHA©) method was created in South Africa in the late 1990s to facilitate the diagnosis of parasitic infection and slow the development of anthelmintic resistance. This method is based on evaluating the color of the eye mucosae membranes of the sheep, correlating with the degree of anemia, and consequently, parasitic infection by *Haemonchus.* As only animals with clinical signs of parasitic infection, especially anemia, receive treatment, the selective use of anthelmintic treatment reduces the risk of developing anthelmintic resistance (Mahieu, 2017; Rosalinski-Moraes et al., 2012).

As it is an easy-to-perform and low-cost method, FAMACHA© is used in several countries, including Brazil; it has demonstrated its efficacy under different bioclimatic

conditions and has become a useful substitute to parasitological tests, such as fecal egg counts (FEC), for the selective treatment of animals with worm diseases (Ferreira et al., 2019; Hupp et al., 2018; Rosalinski-Moraes et al., 2012; Quirino, Carneiro-Silva, Costa, & Madella-Oliveira, 2011). However, its efficacy in the Brazilian Cerrado is still unknown. Thus, this study aimed to evaluate the FAMACHA© method as an alternative tool to diagnose of *haemonchosis* in sheep reared in the Brazilian Cerrado based on the sensitivity and specificity, as well as to determine the clinical signs of Haemonchus sp. infection in sheep. The correlation with packed cell volume (PCV) and FEC was also analyzed.

Material and Methods

Ethics statement

This study was approved by the Ethics Committee on the Use of Animals of the University of Brasília (Brasília, DF, Brazil) under process No. 70/2017.

Clinical and parasitological evaluation of animals

In the Distrito Federal, a region with bioclimatic conditions typical of the Brazilian Cerrado, sheep from seven farms with a semi-intensive rearing system were subjected to clinical and parasitological evaluations. During a period of two years (from December 2017 to July 2019), sheep representing at least 10% of the studied flocks were evaluated. A total of 1,435 sheep was selected by systematic random sampling and grouped into five production categories: pregnant ewes (n=388), lactating ewes (n=218), non-pregnant/lactating ewes (n=378), breeding males (n=113), and weaned lambs (n=338). The sheep were also grouped according to their breed as crossbred sheep (n=759), Santa Inês (n=365), Dorper (n=186), White Dorper (n=70), Île de France (n=35) and Suffolk (n=20).

The sheep were clinically evaluated using the FAMACHA© method (Van Wyk & Bath, 2002). To determine the PCV, blood samples were collected by jugular venipuncture using vacuum tubes containing EDTA anticoagulant (Lopes, Biondo, & Santos, 2007). For coproparasitological evaluation, feces samples were collected directly from the rectal ampulla at dawn, adequately conditioned in separate plastic bags, and stored in a refrigerated Styrofoam container. Subsequently, the FEC of Strongylidae helminths was determined using the modified Gordon and Whitlock technique (Chagas, Niciura, & Molento, 2011; Ueno & Gonçalves, 1998), and coproculture was performed using the Roberts & O'Sullivan technique (Ueno & Gonçalves, 1998). The morphometric and morphological classification of infective larvae (L3) were based on the identification keys (Ueno & Gonçalves, 1998; Van Wyk, Cabaret, & Michel, 2004). FEC analyses were done individually for each sample, while coprocultures were processed as pool samples from animals of the same categories.

Associating FEC, PCV and FAMACHA© exams for diagnosis of sick animals, sheep clinical signs of *Haemonchus* infection were determined. Sensitive animals were considered with FEC values ≥ 800 (Hansen & Perry, 1994; Molento, Braz, & Kloster, 2015), PCV values < 24% (reference values: 24 to 40%; Viana, 2007), FAMACHA© score ≥ 3 (Van Wyk, Malan, & Bath, 1997; Van Wyk & Bath,

2002). Animals were classified as resilient, or tolerant, when they presented average FEC values ≥ 800, PCV values within the reference values for the species, in addition to FAMACHA© 1-3 score. The animals that presented FEC values < 800, PCV within the reference values, in addition to FAMACHA© 1-3 score, were classified as resistant to *Haemonchus* sp. Animals that did not fit these three parameters previously described, didn't receive a clinical classification.

Statistical analyses

In order to calculate the sensitivity, specificity, and positive (PPV) and negative predictive values (NPV) of the FAMACHA© method, PCV was used as the gold standard test for comparison, according to Vatta et al. (2001). A cross-table between PCV (categorized into two classes, sick or healthy, based on two cutoff points, 18% or 23%) and FAMACHA© (categorized into two classes, sick or healthy, based on FAMACHA© scores, where animals with FAMACHA© score 4 and 5, or 3, 4 and 5 were considered sick, and animals with FAMACHA© 1, 2 and 3, or 1 and 2, were considered healthy, respectively) was made (Table 1). The respective cutoff points were chosen based on the limit values of PCV for animals classified with FAMACHA© score 3 (PCV ≥18% e ≤22%). Therefore, the two variables were associeated and were analysed for their ability to classify diseased animals (low PCV) as true positive and healthy animals (high PCV) as true negatives. The diagnostic parameters were evaluated in animals with PCV < 18% and FAMACHA© scores of 4 and 5, and PCV < 23% and FAMACHA© scores of 3, 4, and 5. The animals were considered healthy when the PCV was ≥ 18% and FAMACHA© scores were 1, 2, or 3, and PCV was ≥ 23% and FAMACHA© scores were 1 and 2. Sensitivity, specificity, PPV, and NPV of FAMACHA© were determined for the different animal production categories and breeds, using the Stata 12 statistical software (StataCorp, 2011). Spearman correlation coefficients between the FAMACHA© scores and PCV, FAMACHA© scores and FEC, and PVC and FEC were calculated using the Jamovi 1.6 statistical software (Jamovi, 2021; R Core Team [R], 2020).

Table 1
Cross-table for determining sensitivity, specificity, and positive (PPV) and negative predictive values (NPV) of the FAMACHA© method on sheep from different breeds reared on Brazilian Cerrado. Packed cell volume (PCV) was categorized into two classes, based on two cutoff points, 18% or 23%. FAMACHA© was categorized into two classes, FAMACHA© scores of 4 and 5, and 3, 4, and 5*

FAMACHA© score	With anemia (PCV <18 or <23%)	Without anemia (PCV ≥18 or ≥23%)
Positive (4, 5 or 3, 4, 5)	Truly Positive (VP)	False Positive (FP)
Negative (1, 2, 3 or 1, 2)	False Negative (FN)	Truly Negative (VN)

Sensitivity = (VP/(VP+FN))x100; Specificity = (VN/(FP+VN))x100; Negative Predictive Value = (VN/(FN+VN))x100; Positive Predictive Value = (VP/VP+FP))x100.
*Adapted from Vatta et al. (2001).

Results and Discussion

In the coproparasitological evaluation, the mean FEC from the evaluated sheep was 1,153.34 ± 2,452.38 eggs/g. Four were the genus of helminths observed in the total coproparasitologicals evaluation, *Haemonchus* spp. (76.4%, [confidence interval] CI 95% 72.5-80.3), *Trichostrongylus* spp. (20.2%, CI 95% 16.5-23.9), *Oesophagostomum* spp. (2.9%, CI 95% 2.0-3.7) and *Cooperia* spp. (0.5%, CI 95% 0.0-0.9). *Haemonchus* larvae was predominant in all production categories (74.6% in pregnant ewes; 84.3% in lactating ewes, 69.6% in non-pregnant/lactating ewes, 73.2% in breeding males and 80.8% in weaned lambs). Approximately 80% of the animals had FAMACHA© scores of 1 (466 animals – 32.5%) or 2 (679 animals – 47.3%), and therefore, no anthelmintic treatment was recommended. Animals with FAMACHA© score 3, 4 or 5, represented 16.7% (239 animals), 3.2% (46 animals) and 0.3% (5 animals), respectively. The PCV and FEC values for the different FAMACHA© scores are listed in Table 2.

Table 2
Packed cell volume (PCV) and fecal egg counts (FEC) for the different FAMACHA© scores on sheep from different breeds reared on Brazilian Cerrado

FAMACHA© Score	PCV (%)			FEC		
	Mean	SD	CI (95%)	Mean	SD	CI (95%)
1	33.8	0.2	[33.4-34.2]	705.8	63.0	[582.1-829.4]
2	30.7	0.2	[30.4 to 31.0]	935,3	56.0	[825.5-1,045.2]
3	27.5	0.3	[26.8-28.1]	1,859.2	219.3	[1,428.9-2,289.4]
4	22.0	0.8	[20.3-23.7]	4,844.6	1,124.5	[2,638.7-7,050.4]
5	14.6	2.2	[10.3-18.9]	4,770.0	1,200.9	[2,414.3-7,125.7]

SD: standard deviation
CI: confidence interval.

The number and proportion of animals truly positive, truly negative, false positive, false negative, and precise diagnostic in different cutoffs for FAMACHA© score and PCV are observed in Table 3. The sensitivity, specificity, PPV, and NPV, considering the PCV cutoff points of < 18% or 23%, and FAMACHA© scores 3, 4, and 5, or 4 and 5, for the total flock, production categories, and breeds evaluated are shown in Tables 4, 5, and 6, respectively. Irrespective of the production categories or breeds, the FAMACHA© showed high sensitivity (70.6%) in evaluating animals with FAMACHA© scores > 3 and PCV < 23%. In evaluating diseased animals with FAMACHA© scores ≥ 4 and PCV < 18%, a high specificity (97.5%) was observed in the method (Table 4).

Table 3
Number and proportion of animals truly positive, truly negative, false positive, false negative, and precise diagnostic in different cutoffs for FAMACHA© score and Packed cell volume (PCV), on sheep from different animal production category and breeds reared on Brazilian Cerrado

	Truly Positive	Truly Negative	False Positive	False Negative	Precise Diagnostic
FAMACHA© score 4 and 5, and PCV <18					
Total animals	16 (1.11%)	1,374 (95.75%)	35 (2.44%)	10 (0.70%)	1,390 (96.86%)
Pregnant Ewes	3 (0.77%)	369 (95.10%)	11 (2.83%)	5 (1.29%)	372 (95.87%)
Lactating Ewes	4 (1.83%)	201 (92.20%)	11 (5.04%)	2 (0.92%)	205 (94.04%)
Non-Pregnant/lactating Ewes	1 (0.26%)	373 (98.68%)	4 (1.06%)	-	374 (98.94%)
Breeding Males	-	113 (100.00%)	-	-	113 (100.00%)
Weaned Lambs	8 (2.37%)	318 (94.08%)	9 (2.66%)	3 (0.88%)	326 (96.45%)
Crossed	11 (1.45%)	718 (94.60%)	24 (3.16%)	6 (0.79%)	729 (96.04%)
Santa Inês	2 (0.54%)	355 (97.26%)	7 (1.92%)	1 (0.27%)	357 (97.80%)
Dorper	2 (1.07%)	181 (97.31%)	2 (1.07%)	1 (0.54%)	183 (98.37%)
White Dorper	-	67 (95.71%)	2 (2.86%)	1 (1.43%)	67 (95.71%)
Suffolk	-	20 (100.00%)	-	-	20 (100.00%)
Ile de France	1 (2.86%)	33 (94.28%)	-	1 (2.86%)	34 (97.14%)
FAMACHA© score 3, 4 and 5, and PCV <23					
Total animals	72 (5.02%)	1,115 (77.70%)	218 (15.19%)	30 (2.09%)	1,187 (82.71%)
Pregnant Ewes	15 (3.86%)	295 (76.03%)	72 (18.56%)	6 (1.55%)	310 (79.89%)
Lactating Ewes	19 (8.71%)	142 (65.14%)	50 (22.93%)	7 (3.21%)	161 (73.85%)
Non-Pregnant/lactating Ewes	10 (2.64%)	313 (82.80%)	46 (12.17%)	9 (2.38%)	323 (85.45%)
Breeding Males	2 (1.77%)	98 (86.72%)	13 (11.50%)	-	100 (88.49%)
Weaned Lambs	26 (7.69%)	267 (79.00%)	37 (10.95%)	8 (2.37%)	293 (86.69%)
Crossed	46 (6.06%)	576 (75.89%)	116 (15.28%)	21 (2.77%)	622 (81.95%)
Santa Inês	12 (3.79%)	283 (77.53%)	64 (17.53%)	6 (1.64%)	295 (80.82%)
Dorper	6 (3.22%)	161 (86.56%)	18 (9.68%)	1 (0.54%)	167 (89.78%)
White Dorper	2 (2.86%)	49 (70.00%)	18 (25.71%)	1 (1.43%)	51 (72.86%)
Suffolk	-	18 (90.00%)	1 (5.00%)	1 (5.00%)	18 (90.00%)
Ile de France	6 (17.4%)	28 (80.00%)	1 (2.86%)	-	34 (97.14%)

Precise diagnostic: truly positive and truly negative.

Table 4
Sensitivity, specificity, positive (PPV) and negative (NPV) predictive values in different FAMACHA© and PCV cutoff points for identification of diseased sheep from different animal production category and breeds reared on Brazilian Cerrado

Positive;Diseased animals	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Scores 4 and 5; PCV <18%	61.5	97.5	31.4	99.3
Scores 3, 4 and 5; PCV <23%	70.6	83.6	24.8	97.4

PCV: packed cell volume.

Table 5
Comparison of different cutoffs for FAMACHA© and packed cell volume (PCV) in the identification of diseased sheep from different breeds in each animal production category

Positive;Diseased animals	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Pregnant Ewes				
Scores 4 and 5; PCV <18%	37.5	97.1	21.4	98.7
Scores 3, 4 and 5; PCV <23%	71.4	80.4	17.2	98.0
Lactating Ewes				
Scores 4 and 5; PCV <18%	66.7	94.8	26.7	99.0
Scores 3, 4 and 5; PCV <23%	73.1	73.9	27.5	95.3
Non-Pregnant/lactating Ewes				
Scores 4 and 5; PCV <18%	100.0	98.9	20.0	100.0
Scores 3, 4 and 5; PCV <23%	52.6	87.2	17.8	97.2
Breeding Males				
Scores 4 and 5; PCV <18%	-	100.0	-	100.0
Scores 3, 4 and 5; PCV <23%	100.0	88.3	13.3	100.0
Weaned Lambs				
Scores 4 and 5; PCV <18%	72.7	97.2	47.0	99.1
Scores 3, 4 and 5; PCV <23%	76.5	87.8	41.3	97.1

PPV: positive predictive value
NPV: negative predictive value.

Table 6
Comparison of different cutoffs for FAMACHA© and packed cell volume (PCV) in the identification of diseased sheep from different breeds reared on Brazilian Cerrado

Positive;Diseased animals	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Crossed				
Scores 4 and 5; PCV <18%	64.7	96.7	31.4	99.2
Scores 3, 4 and 5; PCV <23%	68.6	83.2	28.4	96.5
Santa Inês				
Scores 4 and 5; PCV <18%	66.7	98.1	22.2	99.7
Scores 3, 4 and 5; PCV <23%	66.7	81.5	15.8	97.9
Dorper				
Scores 4 and 5; PCV <18%	66.7	98.9	50.0	99.4
Scores 3, 4 and 5; PCV <23%	85.7	89.9	25.0	99.4
White Dorper				
Scores 4 and 5; PCV <18%	-	97.1	-	98.5
Scores 3, 4 and 5; PCV <23%	66.7	73.1	10.0	98.0
Suffolk				
Scores 4 and 5; PCV <18%	-	100.0	-	100.0
Scores 3, 4 and 5; PCV <23%	-	94.7	-	94.7
Ile de France				
Scores 4 and 5; PCV <18%	50.0	100.0	100.0	97.0
Scores 3, 4 and 5; PCV <23%	100.0	96.5	85.7	100.0

PPV: positive predictive value
NPV: negative predictive value.

The FAMACHA© is a valuable tool used to identify gastrointestinal parasitic infection in animals with signs of *Haemonchus* sp. infection (Veríssimo et al., 2012; Amarante, Silva, & Ragozo, 2014). As observed in this study, of the 1,435 sheep evaluated, 20.2% showed a FAMACHA© score indicative of mucosal pallor (scores 3, 4, or 5) and consequently, possible parasitic infection by *Haemonchus* sp., the predominant helminth observed in Distrito Federal, Midwestern Brazil. Considering FAMACHA© scores ≥ 4 as indicative for treatment, only 3.5% of the animals received recommendation of anthelmintic treatment.

A frequent problem with the FAMACHA© method is the treatment choice to be adopted for sheep with a score of 3. According to the FAMACHA© method and chart score, a score of 3 is considered borderline; therefore, the need for anthelmintic treatment is uncertain. The adoption of score 3 as indicative of requiring anthelmintic treatment increased the sensitivity of the method and reduced the specificity. This observation corroborated previous results, which also observed a high specificity when considering a score of 4 and PCV < 18% as the cutoff point and an increased sensitivity of the method (from 61.5% to 70.6%) when a score of 3 was considered

as the cutoff point. Oliveira et al. (2012) and Fernandes et al. (2015) also observed higher sensitivity when considering diseased animals with a FAMACHA© score of 3. Maia, Rosalinski-Moraes, Torres-Acosta, Cintra and Sotomaior (2015) observed 100% sensitivity in the FAMACHA© method, regardless of the score. According to the authors, only the specificity decreased, from 87.8% to 62.4%, when treating individuals with a score of 3.

The increase in the diagnostic sensitivity of the FAMACHA© method based on considering animals classified with score 3 as sick was linked decreased PPV. The mean value of PCV in animals with FAMACHA© score 3 were within the reference value for sheep, which may justify the low PPV. Such observation leads to a greater possibility of treating false positive animals. Even so, the proportion of false negative animals, remained below 5%, considering the cutoff 3 of the FAMACHA© test. Therefore, the discriminative character of the test for diagnosing sick animals proved to be satisfactory. Considering the health impact of Haemonchus sp. infection, higher sensitivity is desired for infected animals at risk of death due to anemia (Oliveira et al., 2012). This fact is especially important in production categories considered more susceptible to helminthic infection, as pregnant, lactating ewes and weaned lambs (Rocha et al., 2011; Amarante et al., 2014).

The FAMACHA© score ≥ 3 increased diagnostic sensitivity in these aforementioned categories, an important finding of the present study, guaranteed treatment of clinically ill animals. In weaned lambs, diagnostic efficacy using exclusively the FAMACHA© method was not proven (Cintra, Ollhoff, Weber, & Sotomaior, 2019). Accordingly, in addition to FAMACHA©, determining weight gain is also an important measure to be adopted for the haemonchosis clinical diagnosis and selective treatment in young animals. Since in the present study the evaluation of weight gain was not determined, the exclusive use of the FAMACHA© method as a diagnostic technique for haemonchosis proved to be an effective alternative for selective treatment, given the high values of sensitivity and specificity observed, as well as the high proportions of accurate diagnosis, regardless of the cutoff point in the determination of sick animals (precise diagnostic from 94.04 until 100% in FAMACHA© 4 and 5 plus PCV <18, and 72.86 until 97.14% in FAMACHA© 3, 4 and 5 plus PCV <23, respectively) in all categories of animal production and breeds evaluated, including weaned lambs. Fernandes et al. (2015) also observed diagnostic efficacy in the FAMACHA© method both in adult sheep and suckling lambs.

Among the animal production categories, there was a broad variability in sensitivity values (37.5% to 100%) among diseased animals with FAMACHA© scores ≥ 4 and PCV < 18%. A lower sensitivity was observed among pregnant ewes, which is an undesirable finding since this category has a high risk for helminthic infections. In contrast, among diseased animals with scores ≥ 3 and PCV < 23%, a higher sensitivity and lower discrepancy between these values were observed in different production categories, except for non-pregnant/lactating ewes, which presented high sensitivity and specificity with FAMACHA© scores of 4 and 5 and PCV > 18%.

Among the different breeds evaluated, the sensitivity and specificity values remained highly similar in the three breeds with higher representativeness (crossbred sheep, Santa Inês, and Dorper). This suggests that the

difference in breeds has a low impact on the method's applicability, given the conditions of this study. Moors and Gauly (2009) observed a difference in mucosal coloration and FAMACHA© score classification in two distinct German breeds (Black Head Mutton and Leine). However, an important factor in that study was the presence of a mixed parasitic infection, without a predominance of *Haemonchus* sp. In Brazilian semiarid region, Ferreira et al. (2019) observed high sensitivity and specificity when evaluating the efficiency of the method in Morada Nova breed.It is worth mentioning that in all the animal production categories and breeds, the specificity and NPV remained high (73.1-100%), as well as false negative proportions (<5%), demonstrating the ability of the method to discriminate healthy from diseased animals in the flock; therefore, it can safely select animals that don't need to be treated. The high sensitivity and specificity of the method directly influences the animal's health by reducing the use of anthelmintics, and consequently, the pressure of selection of resistant parasites, allowing the maintenance of susceptible helminths (Torres-Acosta, Mendoza-de-Gives, Aquilar-Caballero, & Cuellar-Ordaz, 2012). Considering that deworming *en masse* is a common practice to control gastrointestinal parasites in sheep production systems in the Brazilian Cerrado (Moreira, Mota, Gonçalves, Rocha, & Borges, 2021), correcting this practice through the implementation and routine use of the FAMACHA© technique will significantly reduce the unnecessary use of anthelmintic drugs in healthy animals. It also lowers costs by reducing the number of treated individuals.

The Spearman correlation coefficients between the variables FEC and PCV was -0.47, between FEC and FAMACHA© score was 0.22, and between PCV and FAMACHA© score was -0.46 ($p<0.001$ for all). The correlation analyses revealed a moderate negative correlation between PCV and FAMACHA© (-0.46 in the total herd flock). This result demonstrates that a low PCV percentage is positively correlated with a high FAMACHA© score, which is in accordance to the test principle (Van Wyk & Bath, 2002). FAMACHA© and FEC showed a low positive correlation (0.22). The low correlation between the variables can be justified by the resilience of some animals that are clinically healthy even with a high parasite worm burden (Amarante, 2015). A moderate negative correlation was observed between FEC and PCV (-0.47). The findings of the descriptive data analyses show that low PCV values and a high mean FEC are positively correlated with high FAMACHA© scores. Similarly, Santa Inês sheep showed negative correlation indices between FAMACHA© and PCV (-0.42) and PCV and FEC (-0.40) and a positive correlation between FAMACHA© and FEC (0.21) (Jiménez-Sanz et al., 2016). The results obtained by Rosalinski-Moraes et al. (2012) also support this finding, with correlation indexes of -0.74 between FAMACHA© and PCV, -0.69 between PCV and FEC, and 0.37 between FEC and FAMACHA©.

Regarding the presented clinical signs, associating diagnostic parameters (FEC, PCV and FAMACHA©), 4.9% of the animals evaluated were sensitive to Haemonchus gastrointestinal parasites, showing anemia associated with high parasitic load and, therefore, requiring anthelmintic treatment. The clinical condition of resilience to helminthic infection was observed in 22% of the animals. The vast majority of the evaluated sheep (54.5%) demonstrated clinical signs of resistance to gastrointestinal parasites, and 18.6% were considered without clinical classification.

Economically, the selective treatment of sheep using the FAMACHA© method allows considerable savings from anthelmintic drug use. Assuming mass treatment, 1,435 sheep evaluated in this study would receive deworming, regardless of whether or not they needed therapeutic intervention. When considering the selection and treatment of animals based on the FAMACHA© method, for animals with scores of 3, 4, and 5, only 20.21% of the animals would have recommended anti-helminthic treatment (290 heads). By the association of FEC, PCV and FAMACHA© exams, the number of animals to be treated is even smaller, with only 4.88% (70 heads) of the sheep classified as sensitive. The selective control may result in 79.8% and 95.1% of saving, respectively, and avoid unnecessary treatment of clinically healthy animals. This targeted treatment reduces the selection pressure for helminthes resistant to anthelmintic drugs and consequently, delays anthelmintic resistance.

Conclusions

In sheep reared in the Brazilian Cerrado, the FAMACHA© method is a valuable diagnostic alternative, given the predominance of *Haemonchus* in the production systems, because of its high sensitivity and specificity. The high proportion of accurate diagnoses of the test, regardless of its cutoff points, makes it an important ancillary tool for the diagnosis of haemonchosis in the region, and consequently in the implementation of selective treatment in the routine of sheep breeding farms. Thus, the FAMACHA© method can be a useful alternative to control *Haemonchus* infection in sheep rearing systems from Brazilian Cerrado.

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