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## Reliability of FAMACHA<sup>®</sup> chart for the evaluation of anaemia in goats in and around Maiduguri

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### Abstract

The reliability of FAMACHA<sup>®</sup> chart for identifying anaemic goats was compared with Packed Cell Volume (PCV). The colour of the lower eyelids was graded with FAMACHA<sup>®</sup> chart based on FAMACHA<sup>®</sup> scores (FS) of 1-5. The animals were scored from severely anaemic (white or FS 5) through moderately anaemic (pink or FS 3) to non-anaemic (red or FS 1). The prevalence of anaemia in the animals was also determined. This study was done to control death of animals from anaemia by the use of FAMACHA<sup>®</sup> chart, a method which does not need laboratory analysis and can be used by the illiterate with minimal training. A total of 415 goats were used for the study. Blood samples were collected for determination of PCV. Data for both FS and PCV were evaluated using two separate cut off points for each sample (values of FS 4 and 5 or FS 5 and PCV values of  $\leq 19\%$  or  $\leq 15\%$ ), to determine anaemic animals. The FAMACHA<sup>®</sup> Score of the evaluated goats was normally distributed, with FS 3 occurring most frequently (34.5%), whilst, the least was FS 1 (5.3%). There was a high negative correlation between FS and PCV ( $r = -0.69$ ,  $P < 0.02$ ). The sensitivity of FS was high (64%) when FS 5 and PCV  $\leq 19\%$ , were used to determine anaemia, but when FS 4&5 and PCV  $\leq 15\%$  were used, the sensitivity decreased to 22%. Thus, it was concluded that FAMACHA<sup>®</sup> scores of 5 gave the most reliable indicator of anaemia in goats, coinciding with the PCV values of  $\leq 19\%$ . A high Likelihood Ratio of 22 was recorded in this study. Indicating that FS will be positive for anaemia 22 times in anaemic animals than in non anaemic animals.

**Keywords:** Anaemia, FAMACHA<sup>®</sup> chart, Goat, Reliability, Sensitivity

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### Introduction

Goats have a wide distribution in Nigeria (Anene *et al.*, 1994). Their ability to survive on native pasture, high reproductive rate, low cost of feeding and requirement for minimal capital input make their production attractive to all classes of farmers (Aliyu, 1990).

In many parts of the world, goats are raised for meat, milk or fibre. In Nigeria however, the primary objective of goat production is meat, especially in the rural areas (Saidu, 1978).

One of the major constraints to goat production in Nigeria is diseases associated with anaemia. A novel and easy system that uses the severity of clinical anaemia to identify animals with severe anaemia is

the FAMACHA<sup>®</sup> (Fafan Malan Chart) system. This system was developed in South Africa and is used to grade the degree of anaemia by assessment of the shades of red/pink in the lower eyelid of animals (Bath *et al.*, 1996). Use of this test has been shown to improve treatment quality and herd health, and to reduce drug costs (Van Wyk & Bath, 2002). The test has been extensively tested in South Africa and more recently in the USA and Caribbean (Kaplan *et al.*, 2004). This study was aimed at determining the reliability of the FAMACHA<sup>®</sup> system to evaluate degree of anaemia compared with PCV of goats in and around Maiduguri.

**Table 1:** Two by two contingency table FAMACHA<sup>®</sup> system

	Anaemic	Not anaemic	Total
Pale mucous Membrane	True Positive (a)	False Positive (b)	a+b
Red mucous Membrane	False Negative (c)	True Negative (d)	c+d
Total	a+c	b+d	a+b+c+d(N)
Sensitivity = a/ a+c	PPV= a/a+b	LR= sensitivity/(1-specificity)	
Specificity = d/ d+b	NPP = d/c+d		

## Materials and methods

### Study area and animals

The study was conducted between August 2005 and July 2007 in and around Maiduguri in Borno State, within the semi-arid zone of north-eastern Nigeria. A total of 415 goats were surveyed at the Maiduguri metropolitan abattoir, the University of Maiduguri research farm and in small holder farms in the study area. The animals were evaluated using the FAMACHA<sup>®</sup> system. The colour of the mucous membrane of the lower eyelid was compared with the FAMACHA<sup>®</sup> system, which is a laminated FAMACHA<sup>®</sup> anaemia guide having five colour charts according to the degree of anaemia. The colour of the ocular mucous membranes of each animal was examined and classified into one of the five categories according to the FAMACHA<sup>®</sup> eye colour chart: 1 = red, non anaemic; 2 = red-pink, non-anaemic; 3 = pink, mildly-anaemic; 4 = pink-white, anaemic; 5 = white, severely anaemic. Blood samples were then collected from the animals for laboratory analysis.

### Statistical analysis

The sensitivity and specificity, the positive and negative predictive values and the likelihood ratio of FAMACHA<sup>®</sup> score were determined using two by two contingency table (table 1). Data from animals were analyzed, Spearman correlation coefficients were calculated (using GraphPAD instat<sup>®</sup> version 3.00) to determine the relationship between eye scores and PCV. Two-way frequency tables with PCV by eye score were created according to Vatta *et al.* (2001). Eye score values of 4 and 5 or 5 were considered anaemic and eye score values of 1, 2, 3 or 1, 2, 3, 4 were considered non-anaemic. Packed cell volume values were considered anaemic if  $\leq 19\%$  or  $\leq 15\%$ . These two levels were used to provide alternative views of the data; since no precise value for PCV has been clearly established at which anaemia crosses a threshold of clinical importance (Kaplan *et al.*, 2004). Sensitivity, specificity were calculated for the data according to Vatta *et al.* (2001). A true positive result was defined as animals



Figure 1: FAMACHA<sup>®</sup> anaemia guide

that were anaemic (PCV  $\leq 15\%$  or  $\leq 19\%$ ) with pale eye scores (5 or 4 and 5). A false positive result was defined as animals that were not anaemic (PCV  $> 15\%$  or  $> 19\%$ ) with pale eye scores. A false negative result was defined as animals that were anaemic with red or pink eye scores (1, 2, 3 or 1, 2, 3, 4). A true negative result was defined as animals that were not anaemic with pink or red eye scores (Vatta *et al.*, 2001)

## Results

The frequency distribution of FAMACHA<sup>®</sup> Score of the evaluated goats was normal with FS 3 having the highest percent of 34.5% as presented in Table 2. There was a high negative correlation between FS and PCV ( $r = -0.69$ ,  $P < 0.02$ ). Table 2: The sensitivity of FS for assigned ranges of PCV in goats when the cut off point of PCV  $\leq 19\%$  and FS of 4 and 5 was considered anaemic is presented in Table 3. Correct interpretation was made in 71.6% of animals while 4.1% were false positives and 24.3% false negatives with sensitivity and specificity of 38% and 93%, having Positive Predictive Value (PPV) of 78.8% and Negative Predictive Value (NPV) of 69.9% respectively. The Likelihood Ratio (LR) was 5.4. However, at the cut-off point of PCV  $\leq 19\%$  and FS of

5 (Table 4), correct interpretation was made in false negatives were 4.3%. The sensitivity and specificity were 64% and 87% respectively. The PPV was 40.0% and NPV 94.6% while the LR was 4.9. Table 5, Presents the sensitivity of FS for assigned ranges of PCV in goats when the cut off point of PCV  $\leq 15\%$  and FS of 4 and 5 were considered anaemic. Correct interpretation was made in 68.4% of animals while 0.7% were false positives and 30.8% false negatives. The sensitivity and specificity were 22%

84.1% of animals. False positives were 11.6% and 99% respectively, while PPV and NPV were 66.0% and 92.3% respectively, with a LR of 22. When the cut off point of PCV  $\leq 15\%$  but only FS of 5 was considered anaemic (Table 6), correct interpretation was made in 90.1% of animals while 3.6% were false positives and 6.3% false negatives with sensitivity and specificity of 48% and 96% respectively. The PPV and NPV were 61.5% and 93.1% respectively whilst the LR was 12.

**Table 2:** The frequency distribution of FAMACHA<sup>®</sup> Score of the animals examined

FAMACHA <sup>®</sup> Score (FS)	Number of animals (%)	PCV (%)
1	22 (5.3)	30.3 $\pm$ 5.3
2	86 (20.7)	29.4 $\pm$ 5.1
3	143 (34.5)	26.0 $\pm$ 5.0
4	114 (27.5)	23.3 $\pm$ 5.7
5	50 (12.1)	19.0 $\pm$ 7.8

**Table 3:** Sensitivity of FAMACHA<sup>®</sup> system in goats with assigned class intervals of PCV where FS of 5 and 4 were considered as anaemic (PCV  $\leq 19$ )

PCV interval	class	Number of animals	FAMACHA <sup>®</sup> score validation by PCV*			
			Diagnosis of Anaemia		Diagnosis of Non Anaemia	
			FS 4-5 True Positive (anaemic)	FS 1-3 False positive (Not anaemic)	FS 4-5 False Negative ( anaemic)	FS 1-3 True Positive (Not anaemic)
9-16		44	38	6	-	-
17-19		36	25	11	-	-
20-32		276	-	-	90	186
33-40		57	-	-	11	46
41-48		2	-	-	0	2
TOTAL		415	63(a)	17(b)	101(c)	234(d)

\* Validation summary: Correct Interpretation = a + d = 63 + 234= 297 (71.6 %), False Positive = b = 17 (4.1 %), False Negative = c = 101 (24.3 %), Sensitivity = 38% ,PPV=78.8%, Specificity =93%, NPV= 69.9%, LR=5.4

**Table 4:** Sensitivity of FAMACHA<sup>®</sup> system in goats with assigned class intervals of PCV where FS of 5 was considered as anaemic (PCV  $\leq 19$ )

PCV class interval	Number of animals	FAMACHA <sup>®</sup> score validation by PCV*			
		Diagnosis of Anaemia		Diagnosis of Non Anaemia	
		FS 5 True Positive (anaemic)	FS 1-4 False Positive (Not anaemic)	FS 5 False Negative ( anaemic)	FS 1-4 True Positive (Not anaemic)
9-16	44	24	20	-	-
17-19	36	8	28	-	-
20-32	276	-	-	14	262
33-40	57	-	-	4	53
41-48	2	-	-	0	2
TOTAL	415	32(a)	48(b)	18(c)	317(d)

\* Validation summary: Correct Interpretation = a + d = 32 + 317= 349 (84.1 %), False Positive = b = 48 (11.6 %), False Negative = c = 18 (4.3 %), Wrong interpretation = b+c = 66 (15.9 %), Sensitivity = 64% ,PPV=40.0%, Specificity =86.9%, NPV= 94.6%  
LR=4.9

**Table 5:** Sensitivity of FAMACHA<sup>®</sup> system in goats with assigned class intervals of PCV where FS of 4 and 5 were considered as anaemic (PCV ≤ 15)

PCV class interval	Number of animals	FAMACHA <sup>®</sup> score validation by PCV*			
		Diagnosis of Anaemia		Diagnosis of Non Anaemia	
		FS 5 True Positive (anaemic)	FS 1-4 False Positive (Not anaemic)	FS 5 False Negative ( anaemic)	FS 1-4 True Positive (Not anaemic)
9-16	44	36	3	-	-
17-19	36	-	-	72	66
20-32	276	-	-	45	134
33-40	57	-	-	11	46
41-48	2	-	-	0	2
TOTAL		36(a)	3(b)	128(c)	248(d)

\* Validation summary: Correct Interpretation = a + d = 36 + 248 = 284 (68.4 %), False Positive = b = 3 (0.7 %), False Negative = c = 128 (30.8 %), Wrong interpretation = b+c = 131 (31.6 %), Sensitivity = 22%, PPV= 92.3%, Specificity = 99%, NPV= 66%  
LR=22

**Table 6:** Sensitivity of FAMACHA<sup>®</sup> system in goats with assigned class intervals of PCV where FS of 5 was considered as anaemic (PCV ≤ 15)

PCV class interval	Number of animals	FAMACHA <sup>®</sup> score validation by PCV*			
		Diagnosis of Anaemia		Diagnosis of Non Anaemia	
		FS4-5 True Positive (anaemic)	FS 1-3 False Positive (Not anaemic)	FS4-5 False Negative (anaemic)	FS 1-3 True Positive (Not anaemic)
9-15	39	24	15	-	-
16-24	138	-	-	14	124
25-32	179	-	-	8	171
33-40	57	-	-	4	53
41-48	2	-	-	0	2
TOTAL		24(a)	15(b)	26(c)	350(d)

\* Validation summary: Correct Interpretation = a + d = 24 + 350 = 374 (90.1 %), False Positive = b = 15 (3.6 %), False Negative = c = 26 (6.3 %), Sensitivity=48%, PPV = 61.5%, Specificity = 96%, NPV= 93.1%, LR=12

## Discussion

The mean PCV concentrations corresponding to individual FS values recorded in the present study were significantly higher compared to those observed by other workers (Vatta *et al.*, 2001; Kaplan *et al.*, 2004; Burke *et al.*, 2007; Miriam *et al.*, 2010). In agreement with previous studies, the distribution of FAMACHA<sup>®</sup> score observed in the current study was normal and the correlation between FS and PCV was high (Kaplan *et al.*, 2004; Burke *et al.*, 2007; Miriam *et al.*, 2010; Spickett *et al.*, 2011).

The choice of PCV and FS cut-off points used to determine FAMACHA<sup>®</sup> chart accuracy for the diagnosis of anaemia in animals has a major effect on test performance, and different cut-offs can be

used depending on whether high sensitivity or high specificity are desired. In this study, FS 4 & 5 and FS 5 were used instead of FS 3, 4, & 5 and FS 4 & 5 used in previous reports (Kaplan *et al.*, 2004; Burke *et al.*, 2007; Miriam *et al.*, 2010). This was because the PCV means of FS 3 observed in the current study were high ( $26.0 \pm 5.0$ ) compared with the PCV value of 18–22% reported by Vatta *et al.* (2001) for the same score in South Africa. This means that based on the study reported by Vatta *et al.* (2001), an animal with PCVs of 18–22% and 13–17% would be scored as FS 3 and 4 whereas in this study these animals would be scored as FS 4 and 5 respectively.

Two different PCV cut-off points of ≤ 19% and ≤ 15% were used in the present study. The lower cut-off

point was selected because the normal range for PCV in goats is 16–45% (Igbokwe *et al.*, 1998) and a PCV of 19% or less is sometimes used as an indication of anaemia in epidemiologic studies, although an animal with a PCV of 19% is not in any immediate health danger (Vatta *et al.*, 2001).

When FS 5 alone and FS 4 & 5 were used as criteria for identifying anaemic animals, correct interpretation was high (68.4-90.1%). When PCV  $\leq$  15% and FS 4 and 5 was used as a criteria for identifying anaemic animals, the results indicated that 0.7% were false positive, suggesting that using PCV  $\leq$  15% and FS 4 and 5 criteria, 0.7% of goats that actually do not need the treatment may end up being treated. On the other hand, a significantly higher false positive of 4.1% was observed when PCV  $\leq$  19% and FS 4 and 5 were used. When PCV  $\leq$  15% and FS 4 and 5 was used, the PPV was 92.3% and sensitivity of 22%. However, when PCV  $\leq$  15% and FS 5 was used, the PPV of 61.5% and sensitivity of 48% was recorded. When the same eye scores criteria (FS 4 and 5) but PCV  $\leq$  19% was used as a cut-off, PPV of 78.8% and sensitivity increased to 38%.

When eye scores of 5 alone was used as criteria for identifying anaemia, the PPV decreased to 40% while sensitivity was best at 64%, but there was a concurrent decrease in specificity. Importantly, the number of false negatives also increased.

When FS 5 was considered as anaemic, the sensitivity of FAMACHA<sup>®</sup> chart was higher than when FS4 & 5 was used at both PCV cut off points. The reverse was however the case for specificity. High sensitivity is more important than high specificity. When anaemic animals are not identified,

it means that the animal can die of anaemia, where as there is no adverse problem if you identify a non anaemic animal as anaemic (Burke *et al.*, 2007; Antonio *et al.*, 2009; Miriam *et al.*, 2010). FS 5 is therefore the most appropriate scores for use in determining anaemia goats in the study area.

The Likelihood Ratio of FAMACHA<sup>®</sup> chart to identifying an anaemic goat is also an important factor to be considered in favour of using the FAMACHA<sup>®</sup> chart in determining anaemia in goats. The LR of 22, indicated that there is likelihood of FS to identify an animal with low PCV to be anaemic 22 times more than animals with high PCV.

In conclusion, this study has shown that the FAMACHA<sup>®</sup> chart is a very useful tool for identifying anaemic goats with a high LR of 22 recorded in and around Maiduguri, located in the semi-arid zone of North-eastern Nigeria. The best sensitivities were recorded when FS 5 and PCV  $\leq$  19% were used to determine anaemia in goats while the best specificity was obtained when FS 4&5 and PCV  $\leq$  15% was used. It is recommended that FAMACHA<sup>®</sup> chart can be used to determine anaemia in goats in and around Maiduguri with scores 4 and 5 considered anaemic. The FAMACHA<sup>®</sup> chart is useful in the field pending when Veterinary laboratory services can be reached.

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