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Macromineral requirements for maintenance and growth in male and female hair sheep

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A better understanding of the nutritional requirements of sheep, especially in terms of minerals, is crucial for improving production. We estimated the net requirements for Ca, P, K, Mg, and Na for gain (NCa_g, NP_g, NK_g, NMg_g, and NNa_g) and maintenance (NCa_m, NP_m, NK_m, NMg_m, and NNa_m) in male and female hair sheep. Six datasets with 248 individual records of hair sheep (139 non-castrated males, 75 castrated males and 34 females) were used to estimate the net macromineral requirements for gain. To estimate the net macromineral requirements for maintenance, 52 observations (26 non-castrated and 26 castrated males) were used. A meta-analytical approach was applied, using non-linear mixed effects models and the study as a random effect. Based on information criteria for model selection, heterogeneous variance functions were more likely to describe mineral requirements with a low level of model selection uncertainty. The adopted criteria allowed the choice of the best models to represent the macromineral requirements. The chosen models explained the observed variability in the sex, and the choices were based on a low level of uncertainty ($w \geq 0.90$). Irrespective of sex, NCa_g and NP_g decreased with increasing BW from 10 to 30 kg and average daily gain (ADG) of 150 g/day, ranging from 1.71–1.38; 1.83–1.57; 1.82–1.51 of Ca and 0.86–0.66; 0.92–0.78; 0.92–0.75 of P for non-castrated males, castrated males, and females, respectively. The NK_g remained constant, with mean values of 0.26 g/day. The NNa_g range was 0.17 to 0.14 g/day for non-castrated males, 0.20 to 0.25 g/day for females, and constant (0.18 g/day) for castrated males with an increase in BW from 10 to 30 kg and an ADG of 150 g/day. Macromineral requirements for maintenance (mg/kg BW) and retention (%) were 23.70 and 54.30 for Ca, 25.33 and 79.80 for P, 11.74 and 5.00 for K, 2.63 and 8.50 for Mg, and 7.01 and 8.10 for Na for males. The International Committees did not provide inferences about the sex influence on mineral requirements. Our study indicates that sex is one factor that influences the macromineral requirements for gain. The information generated in this study can be used to optimize the mineral management of hair sheep in the growing phase in tropical regions.

KEYWORDS

minerals, models, retention, sex, sheep

1. Introduction

Hair sheep are still important animals in many regions of the world, especially in developing countries, where the demand for food products increases with population growth and associated needs. Although the FAOSTAT (1) has not numerically presented the growth rate of sheep in recent years, it reports that the population of small ruminants remains significant.

A better understanding of the nutritional requirements of sheep, especially in terms of minerals, is crucial for improving production. Minerals are essential elements and perform specific functions in the animal's body, accounting for 3.5% of the animal's body weight (2). Information on mineral requirements for growth (3, 4) and maintenance (5) are important to achieve the maximum productive potential of a herd (6). The net maintenance requirement represents the amount of mineral needed to replace endogenous losses (7), while the gain requirement represents the amount of mineral deposited in the animal's body, usually expressed in g or mg per kg of live weight gained or retained protein (8).

The mineral requirements of growing hair sheep is important, as it allows the reduction of costs in diet formulations in addition to reducing the environmental impact due to mineral contamination. Factors inherent to feed, bioavailability, and chemical form of the element, along with aspects related to inter-associations (antagonism and agonism) among minerals, breed or genetic group, and growth stage (9) can also influence dietary requirements.

Most nutritional requirement guidelines establish net mineral requirements for sheep during growing, however, variation in net mineral requirements across sexes has not been studied. Thus, there is a lack about sex effects on mineral requirements, and the International Committees (10–12) do not consider them in their recommendations. The sex is involved in animal physiology and metabolism and has effects on growth, body composition, and productive and reproductive functions. As with protein and energy, the sex modulates mineral dynamics and, in consequence, mineral deposition in the animal's body (13). Thus, the aim of this study was to estimate the net requirements for gain of Ca, P, K, Mg, Na (NCa_g , NP_g , NK_g , NMg_g , and NNa_g) and maintenance (NCa_m , NP_m , NK_m , NMg_m , and NNa_m) in male and female hair sheep. Thus, we hypothesized that the macromineral requirements of hair sheep differ between sexes.

2. Materials and methods

2.1. Database

To determine macromineral requirements for weight gain, the database consisted of six studies (5, 14–18), comprising a total of 248 animals, with 74 animals belonging to the reference group and 174 to the experimental groups and four breeds (Santa Ines, Morada Nova, Brazilian Somali and crossbreed) with three sex classes: non-castrated ($n = 139$), castrated ($n = 75$) males, and females ($n = 34$) (Table 1).

To estimate maintenance requirements, two studies were used (5, 17), comprising a total of 52 observations, with two sexes: non-castrated ($n = 26$) and castrated ($n = 26$) males. The descriptive analyses of the variables used in this study are provided in Tables 2, 3. For the meta-analytical approach, only studies containing information on individual animals fed at maintenance levels (12, 19) and at least two levels above maintenance were included. Furthermore, the studies were conducted with hair sheep raised in tropical regions and contained the following quantitative data on each animal: (i) body weight (BW); (ii) empty BW (EBW); and (iii) mineral body composition based on chemical analysis. All studies used the comparative slaughter method, in which the animals designated as the reference group were slaughtered at the beginning

of the trial in order to estimate the initial body composition of those who remained in the study.

2.2. Chemical analyses

The EBW and mineral composition of reference animals (Supplementary Table S1) were used to estimate the initial EBW, and the mineral body composition of the remaining animals slaughtered at the end of the trials, within each trial. The macromineral retention was calculated as the final mineral content minus the initial mineral content estimated from the reference animals.

The body components (i.e., blood, hide, hooves, head, organs, and right half of the carcass) were homogenized, sampled, and placed in a forced ventilation oven at 55°C for 72 h. Posteriorly, these samples were defatted by extraction in a Soxhlet apparatus for 12 h [(20); method 920.39]. Subsequently, the fat-free body components samples were ground in a ball mill and analyzed for DM [(20); method 967.03], ash [(20); method 942.05], and CP [(20); method 981.10] levels. The body water content was determined as 100% minus the body component DM. Samples of rations, feed, orts, and body components of each study were analyzed for mineral composition through digestion in nitroperchloric acid, according to method INCT-CA M-004/1 (21). The Ca and Mg were determined by adding strontium chloride and read using atomic absorption spectrometry [GBC Avanta Sigma, Hampshire, USA; (22); method 968.08]. The P was determined by reducing the phosphorus-molybdate complex with ascorbic acid, followed by reading with a colorimetric spectrophotometer [(22); method 965.17]. The Na and K concentrations were determined by flame emission spectrometry [Corning 400, NY, USA; (22); method 985.35]. Further details about slaughter procedures, experimental diets, and sample collection of body components can be found in each study (5, 14–18).

2.3. Models and calculations

2.3.1. Net macromineral requirements for gain

To estimate the requirements for gain, initially, the body mineral composition in the EBW was estimated by using the Brody (23) model, as shown in Equation 1:

$$RM = \beta_0 \times EBW^{\beta_1} \quad (1)$$

where RM = retained mineral (mg/kg EBW/day); EBW = empty BW (kg); β_0 and β_1 are parameters of the equation. Thus, the net macromineral requirements for gain were estimated by first derivation of Equation 1, as follows in Equation 2:

$$NRg = EBWG \times (\beta_0 \times \beta_1 \times EBW^{(\beta_1-1)}) \quad (2)$$

where NRg = net mineral requirements for weight gain (g/day); EBW = empty BW (kg); EBWG = empty BW gain (kg); β_0 and β_1 are parameters of the equation.

2.3.2. Net macromineral requirements for maintenance

A linear regression generated from the relationship between retained minerals (mg/kg BW) against mineral intake (mg/kg BW)

TABLE 1 Summary of studies used to estimation of net mineral requirements for the maintenance and gain of hair sheep.

Study	<i>n</i>	Breed	Sex class	BW (kg)	EBW (kg)
Cabral et al. (14)	24	Santa Ines	Castrated	17.90–24.40	13.13–22.65
Pereira et al. (15)	40	Morada Nova	Non-castrated	10.12–31.62	6.58–27.10
Silva et al. (16)	19	Crossbreed	Non-castrated	10.64–33.60	9.77–30.10
	17		Castrated	7.97–31.70	7.68–29.44
	18		Female	7.98–29.40	7.68–26.28
Pereira et al. (18)	47	Brazilian Somali	Non-castrated	10.90–35.74	7.95–31.18
Pereira et al. (5) ^a	14	Morada Nova	Non-castrated	13.66–38.28	9.62–29.38
	16		castrated	12.54–31.25	8.66–23.08
	16		Female	12.74–26.95	8.89–21.71
Pereira et al. (17) ^a	19	Santa Ines	Non-castrated	11.20–33.20	8.30–24.57
	18		Castrated	12.60–31.56	8.50–29.30

^aStudies used in maintenance; BW, body weight; EBW, empty BW.

TABLE 2 Descriptive statistics of the data used to estimate the net requirements of macrominerals for gain in hair sheep.

Item	<i>n</i> ^a	Mean	SD	Maximum	Minimum
Non-castrated males					
BW (kg)	139	21.56	6.54	38.28	10.12
EBW (kg)	139	17.23	5.93	31.18	6.58
Macromineral retention (g)					
Ca	139	172.63	83.98	542.23	42.76
P	139	114.32	61.00	390.74	33.70
K	139	31.34	17.03	88.81	1.56
Mg	139	6.16	4.36	31.59	1.82
Na	139	25.22	14.93	77.29	2.02
Castrated males					
BW (kg)	75	21.50	5.76	31.70	7.97
EBW (kg)	75	16.81	4.87	29.44	7.68
Macromineral retention (g)					
Ca	75	235.45	75.02	395.48	89.33
P	75	133.13	53.59	268.26	55.49
K	75	33.14	19.08	69.74	2.42
Mg	51	6.16	4.36	31.59	1.82
Na	75	22.38	11.93	49.17	2.42
Female					
BW (kg)	34	18.91	5.36	29.40	7.98
EBW (kg)	34	16.03	4.99	26.28	7.68
Macromineral retention (g)					
Ca	34	216.90	85.68	547.40	92.96
P	34	144.56	64.36	394.93	66.57
K	34	20.85	19.16	60.80	2.64
Mg	34	6.97	6.27	34.99	2.34
Na	34	16.23	14.04	52.30	2.69

^a*n*, number of records; BW, body weight; EBW, empty body weight; SD, standard deviation.

TABLE 3 Descriptive statistics of the data used to estimate the net requirements of macrominerals for maintenance in hair sheep.

Item	<i>n</i> ^a	Mean	SD	Maximum	Minimum
Non-castrated males					
BW (kg)	26	25.28	6.00	38.20	16.46
EBW (kg)	26	19.00	4.96	29.28	11.59
Macromineral intake (mg/day)					
Ca	26	142.28	41.73	220.01	77.31
P	26	90.22	23.92	140.69	59.67
K	26	441.27	93.75	575.28	278.37
Mg	26	63.92	16.65	94.78	37.55
Na	26	169.60	38.05	225.04	105.72
Macromineral retention (mg/day)					
Ca	26	62.40	33.06	124.92	10.31
P	26	47.45	20.58	87.97	11.76
K	26	11.39	5.33	19.43	0.09
Mg	26	2.50	1.91	7.27	0.04
Na	26	6.85	4.03	13.35	-0.17
Castrated males					
BW	26	23.09	5.13	31.56	15.98
EBW	26	17.47	4.38	25.15	11.37
Macromineral intake (mg/day)					
Ca	26	143.53	44.89	232.47	77.44
P	26	89.46	25.14	149.78	59.05
K	26	441.32	100.29	622.32	278.84
Mg	26	63.24	16.35	92.99	37.62
Na	26	171.68	43.68	255.66	105.91
Macromineral retention (mg/day)					
Ca	26	48.72	27.00	98.05	2.99
P	26	36.06	22.19	72.05	2.87
K	26	9.39	5.16	18.16	0.97
Mg	26	3.08	2.50	8.90	-0.31
Na	26	6.46	3.65	12.78	1.02

^a*n*, number of records; BW, body weight; EBW, empty body weight; SD, standard deviation.

was used to calculate the net macromineral requirements for maintenance (mg/kg BW), according to Equation 3:

$$RM = \beta_0 + \beta_1 \times MI \quad (3)$$

where, RM = retained mineral (mg/kg BW/day); MI = mineral intake (mg/kg BW/day); β_0 and β_1 are regression parameters, β_0 is considered the net requirement of each mineral for maintenance (mg/kg BW/day), and β_1 is the slope, considered the retention coefficient (%). However, only studies of Pereira et al. (5, 17) were considered to estimate maintenance requirements (Table 2). The estimated requirements based on the EBW were converted to the BW using the factor (1.23) derived from the relationship between BW/EBW (24). The dietary requirements were calculated as a sum of the net requirements for maintenance and gain divided by the retention coefficient.

2.4. Statistical analysis

2.4.1. Model adjustments for gain

The allometric model was applied to predict the retention (mg/kg EBW/day) of macrominerals in relation to EBW (kg) (Equation 4):

$$RM_Y = \alpha X^\beta \quad (4)$$

$$RM_{Y_i} = \alpha_i X_i^{\beta_i} \quad (5)$$

The model presented in Equation 5 differs from Equation 4 because the prediction of mean body retention of the Y-th ($Y = 1, 2, \dots, 5$) mineral (RM_{Y_i}) was separated by the i-th non-castrated males, castrated males, and females, that is, $i = 1, 2$, and 3. Similarly, the estimates of the parameters α_i and β_i represent estimated values separated by i-sex.

2.4.2. Model adjustments for maintenance

The linear model, Equation 6, was applied for predictions of the macromineral requirements for maintenance in non-castrated and castrated males.

$$RM_Y = \alpha + \beta X \quad (6)$$

The unit of measurement of RMy was according to the Y-th ($Y = 1, 2, \dots, 5$) mineral. The parameters β_0 and β_1 are the intercept and slope, respectively.

2.4.3. Description of variance functions and combinations

The variance (σ_Y^2) was also modeled to test its homoscedasticity assumption. In this case, three variance functions were tested, one homogeneous (homoscedastic) and two heterogeneous functions:

$$\sigma_Y^2 = \sigma^2 \quad (7)$$

$$\sigma_Y^2 = \sigma^2 \exp(\rho X) \quad (8)$$

$$\sigma_Y^2 = \sigma^2 |RM_Y|^{2\phi} \quad (9)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 \quad (10)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 \exp(\rho_i X_i) \quad (11)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 |RM_{Y_i}|^{2\phi} \quad (12)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 \exp(\rho_i X_i) \quad (13)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 |RM_{Y_i}|^{2\phi_i} \quad (14)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 \exp(\rho_i X_i) \quad (15)$$

$$\sigma_{Y_i}^2 = \sigma_i^2 |RM_{Y_i}|^{2\phi_i} \quad (16)$$

The parameter σ refers to the standard deviation, and the term σ^2 refers to the random error variance; thus, Equation 7 represents the homogeneous variance with the assumption of homoscedasticity (25, 26). In this case, the homogeneous (Equations 7 and 10), exponential (Equations 8, 11, 13, and 15), and Power-of-the-Means (Equations 9, 12, 14, and 16) variance functions were tested, as they are the most commonly used (27–29). An attempt to accommodate heteroscedasticity was performed with Equations 8 to 16. Due to the possible scaling effect on the variability, two functions (Equations 8 and 9) were used to adjust for the possible scaling effect on the variance (26). The exponential function represented by Equation 8 assumes an exponential increase in the variance σ^2 with an increasing rate (“ ρ ”) as a function of the EBW, represented by the term X. Equation 9 is the Power-of-the-Means Function for the absolute value of RM_Y , with a scaling parameter ϕ . Both the parameters ρ and ϕ are dimensionless, with values ranging from $-\infty$ to ∞ .

For Equations 7–9, a single variance (σ_Y^2) was estimated independent of the i-th sex. Equations 8–16 use heterogeneous variance functions as they describe a non-linear behavior, except for Equation 10. In Equation 10, the heterogeneous behavior was attributed to the σ_i^2 being estimated for the i-th sex with homogeneous variance. Regarding Equations 11, 12, the adjustments occurred with a distinct σ^2 for each i-sex, whereas the parameters ρ and ϕ were common for the database. Contrary to the previous equations, in Equations 13 and 14, the individualized variance function parameters by sex were ρ_i and ϕ_i , with σ^2 being common. The most complex variance functions are described in Equations

15 and 16 as all their parameters are individualized by sex; σ_i^2 , ρ_i (Equation 15), and ϕ_i (Equation 16).

As Equation 4 was combined with the variance functions (Equations 7–9), three models were fitted to the entire dataset of a given mineral, with no sex effect. On the other hand, Equation 5 was combined with Equations 7–16, totaling 10 different combinations. Initially, adding the combinations made with Equations 4 and 5, 13 combinations were made. Subsequently, all these 13 combinations were performed again, but we added the effect of studies on the parameters “ α ” and “ β ” (Equation 4) and α_i and β_i (Equation 5). Finally, 26 combinations were performed for the body retention of each analyzed mineral. The linear model, Equation 6, was combined with the variance functions demonstrated in Equations 7–9, with three models being fitted to each mineral, with no sex effect.

The fits of the model combinations, both retention and maintenance were performed by the procedure for mixed non-linear models – NLMIXED of SAS. The algorithm used to estimate the maximum likelihood method was the Newton–Raphson (tech = NEWRAP). The assumption of normality was assumed and therefore Y, and the covariance between the measurements was considered null since these were not taken in the same experimental unit. The corrected Akaike information criterion (AICc), the variation (Δ) between each model used and the model with the smallest AICc, the model probability (w), and the evidence ratio (ER) were calculated for model evaluation. Twelve predictions of mineral retention were performed between 8 and 30 kg of EBW (Equation 17) by the derivative of Equation 4 (29):

$$\frac{dRM_Y}{dX} = \widehat{\alpha\beta X^{\beta-1}} \quad (17)$$

The mean predictions of retention rates and their respective 95% confidence intervals (95% CI; Equation 18) were estimated from the best fits for each mineral evaluated. The 95% confidence intervals (95% CI; Equation 18) were also estimated for the parameters “ α ” and “ β ” (Equation 4) and α_i and β_i (Equation 5):

$$95\% \text{ CI} = \text{mean} \pm t_{(1-\alpha, df)} SE \quad (18)$$

The term mean refers to the average prediction of the model as well as the parameters. The expression $\pm t_{(1-\alpha, df)}$ SE represents the estimate of the critical value of the two-tailed distribution of the Student’s *t*-test using the significance level $\alpha = 0.05$ and degree of freedom ($df = n - \theta$), where n represents the number of observations and θ corresponds to the number of parameters of the allometric model (Equations 4 and 5) as well as the variance functions represented in Equations 7–16.

3. Results

Combining Equations 4 and 5 with the variance functions described by Equations 7–16 enabled the estimation of the requirements for gain of NCa_g , NP_g , NMg_g , and NNa_g for castrated males, non-castrated males and females. A general equation was generated for NK_g (Table 4). The adopted criteria allowed the choice of the best models to represent the

TABLE 4 Models for estimating body composition and net requirements for gain of hair sheep.

Macrominerals	Sex	Model	Variance parameters	Net requirements for gain ^a (g/day)
Ca	Non-castrated males	20.41 (4.31) $EBW^{0.83}$ (0.10)	$\sigma = 21.14; \rho = 0.06$	$EBWG [(17.03) EBW^{-0.16}]$
	Castrated males	19.02 (4.29) $EBW^{0.88}$ (0.10)	$\sigma = 21.14; \rho = 0.05$	$EBWG [(16.75) EBW^{-0.12}]$
	Female	20.53 (4.36) $EBW^{0.85}$ (0.11)	$\sigma = 21.14; \rho = 0.09$	$EBWG [(17.52) EBW^{-0.14}]$
P	Non-castrated males	11.57 (1.31) $EBW^{0.79}$ (0.06)	$\sigma = 4.69; \rho = 0.14$	$EBWG [(9.19) EBW^{-0.20}]$
	Castrated males	9.96 (1.40) $EBW^{0.86}$ (0.07)	$\sigma = 4.69; \rho = 0.12$	$EBWG [(8.65) EBW^{-0.13}]$
	Female	10.94 (1.55) $EBW^{0.83}$ (0.08)	$\sigma = 4.69; \rho = 0.22$	$EBWG [(9.16) EBW^{-0.16}]$
K	All sex	1.98 (0.52) $EBW^{0.99}$ (0.10)	$\sigma = 1.07; \varphi = 0.47$	$EBWG [(1.96) EBW^{-0.01}]$
Mg	Non-castrated males	0.49 (0.08) $EBW^{0.91}$ (0.12)	$\sigma = 0.24; \varphi = 0.91$	$EBWG [(0.45) EBW^{-0.08}]$
	Castrated males	0.37 (0.08) $EBW^{1.04}$ (0.12)	$\sigma = 0.12; \varphi = 1.18$	$EBWG [(0.39) EBW^{0.04}]$
	Female	0.48 (0.09) $EBW^{0.935}$ (0.13)	$\sigma = 0.05; \varphi = 1.88$	$EBWG [(0.45) EBW^{-0.06}]$
Na	Non-castrated males	1.99 (0.34) $EBW^{0.83}$ (0.09)	$\sigma = 1.05; \varphi = 0.52$	$EBWG [(1.67) EBW^{-0.16}]$
	Castrated males	1.19 (0.18) $EBW^{1.031}$ (0.08)	$\sigma = 1.05; \varphi = 0.23$	$EBWG [(1.23) EBW^{0.03}]$
	Female	0.87 (0.23) $EBW^{1.18}$ (0.10)	$\sigma = 1.05; \varphi = 0.246$	$EBWG [(1.03) EBW^{0.18}]$

^aThe empty body weight gain (EBWG g/day) was calculated according to Herbster et al. (24); EBW, empty body weight.

TABLE 5 Information criteria of fitted models to estimate the net requirements for gain of hair sheep.

Macrominerals	Model	Variance functions	Random effect	AICc	Δ	w	ER	Θ
Ca	Equation (5)	Equation (12)	$\alpha_i; \beta_i$	2563.90	0.00	0.32	1.00	16.00
	Equation (5)	Equation (15)	$\alpha_i; \beta_i$	2564.40	0.50	0.25	1.28	18.00
	Equation (5)	Equation (10)	$\alpha_i; \beta_i$	2564.80	0.90	0.20	1.57	16.00
	Equation (5)	Equation (7)	$\alpha_i; \beta_i$	2566.60	2.70	0.08	3.86	14.00
	Equation (4)	Equation (7)	$\alpha; \beta$	2566.90	3.00	0.07	4.48	6.00
P	Equation (5)	Equation (12)	$\alpha_i; \beta_i$	2167.40	0.00	1.00	1.00	16.00
K	Equation (4)	Equation (8)	$\alpha; \beta$	1551.10	0.00	0.92	1.00	6.00
Mg	Equation (5)	Equation (11)	$\alpha_i; \beta_i$	779.70	0.00	0.98	1.00	16.00
Na	Equation (5)	Equation (13)	$\alpha_i; \beta_i$	1391.30	0.00	0.80	1.00	16.00
	Equation (5)	Equation (11)	$\alpha_i; \beta_i$	1394.20	2.90	0.19	4.26	16.00

AICc, Akaike information criterion corrected for small samples; Δ , Akaike difference; w, model probability; ER, evidence ratio computed for each model; Θ , number of parameters of the allometric model.

requirements for gain, explaining the observed variability among sexes (Table 5).

Irrespective of sex, NCa_g and NP_g decreased with increasing BW from 10 to 30 kg and average daily gain (ADG) of 150 g/day, ranging from 1.71–1.38; 1.83–1.57; 1.82–1.51 of Ca and 0.86–0.66; 0.92–0.78; 0.92–0.75 of P for non-castrated males, castrated males, and females, respectively. The NMg_g independent of BW and gain remained constant for each sex. Females during growth presented greater requirements than non-castrated males for Ca, P, Mg and Na. The NK_g remained constant, with mean values of 0.26 g/day. The NNa_g range was 0.17–0.14 g/day for non-castrated males, 0.20–0.25 g/day for females, and constant (0.18 g/day) for castrated males with an increase in BW from 10 to 30 kg and an ADG of 150 g/day (Table 6).

The Figure 1 shows the amount of water, protein and ash in relation to BW increased from 10 to 30 kg. In the respective weight range, when the 95% confidence limits of NCa_g , NP_g , NMg_g ,

NNa_g , and NK_g were evaluated, uncertainty irrespective of sex increased as BW increased. However, when the NMg_g was evaluated, uncertainty for females was intermediary compared to non-castrated and castrated males.

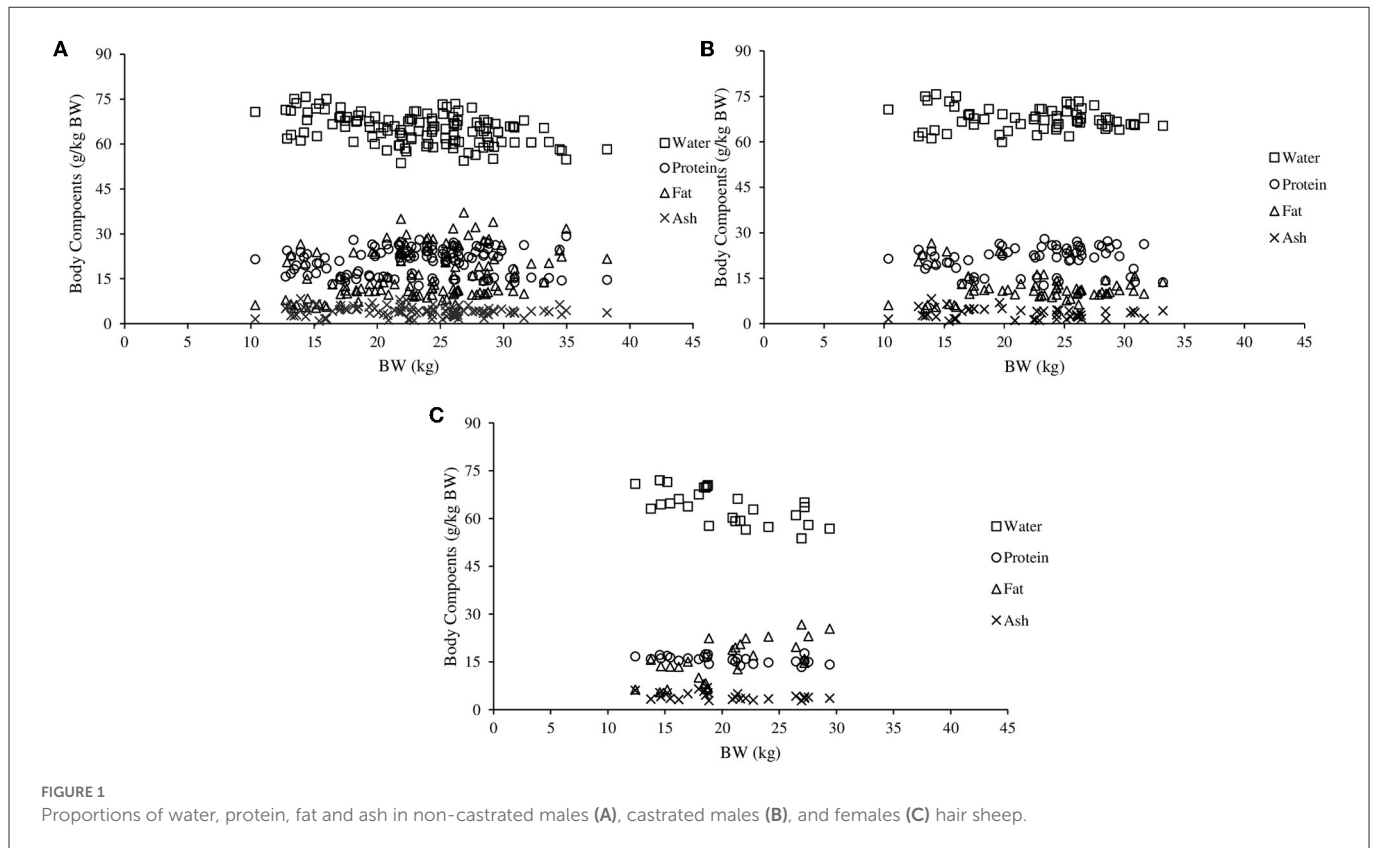
Macromineral requirements for maintenance were estimated by the relationships between minerals retained and mineral intake, combining the linear model (Equation 6) with the variance functions described by Equations 7–9. Sex did not affect the intercept and slope of the linear equations to estimate the Ca (Figure 2A), P (Figure 2B), K (Figure 2C), Mg (Figure 2D), and Na (Figure 2E), therefore, linear equations were generated for each macromineral. The best model to represent the mineral requirements for maintenance was based on the adopted criteria shown in Table 7.

The net mineral requirement for maintenance and retention were 23.70 mg/kg and 54.30% for Ca, 25.33 mg/kg and 79.80% for P, 11.74 mg/kg and 5.00% for K, 2.63 mg/kg and 8.50% for Mg, 7.01

TABLE 6 Net mineral requirements for gain of hair sheep with respective confidence intervals.

BW (kg) ADG (g/day)	10			20			30		
	100	150	200	100	150	200	100	150	200
Non-castrated males									
Ca	1.14 ± 0.64	1.71 ± 0.96	2.28 ± 1.28	0.99 ± 0.77	1.49 ± 1.15	1.99 ± 1.54	0.92 ± 0.83	1.38 ± 1.24	1.84 ± 1.66
P	0.57 ± 0.24	0.86 ± 0.36	1.14 ± 0.48	0.48 ± 0.27	0.72 ± 0.40	0.96 ± 0.54	0.44 ± 0.28	0.66 ± 0.42	0.88 ± 0.56
Mg	0.03 ± 0.03	0.05 ± 0.05	0.07 ± 0.07	0.03 ± 0.04	0.05 ± 0.06	0.07 ± 0.08	0.03 ± 0.04	0.05 ± 0.07	0.06 ± 0.09
Na	0.11 ± 0.09	0.17 ± 0.13	0.22 ± 0.18	0.10 ± 0.09	0.15 ± 0.14	0.20 ± 0.19	0.09 ± 0.10	0.14 ± 0.15	0.18 ± 0.20
Castrated males									
Ca	1.22 ± 0.65	1.83 ± 0.98	2.44 ± 1.30	1.10 ± 0.82	1.66 ± 1.23	2.21 ± 1.64	1.05 ± 0.90	1.57 ± 1.36	2.09 ± 1.81
P	0.62 ± 0.24	0.92 ± 0.36	1.23 ± 0.48	0.55 ± 0.29	0.83 ± 0.44	1.10 ± 0.59	0.52 ± 0.32	0.78 ± 0.48	1.04 ± 0.64
Mg	0.04 ± 0.03	0.06 ± 0.05	0.08 ± 0.07	0.04 ± 0.05	0.06 ± 0.07	0.08 ± 0.09	0.04 ± 0.05	0.06 ± 0.08	0.08 ± 0.11
Na	0.12 ± 0.10	0.18 ± 0.16	0.24 ± 0.21	0.12 ± 0.12	0.18 ± 0.19	0.24 ± 0.25	0.12 ± 0.14	0.18 ± 0.21	0.25 ± 0.28
Females									
Ca	1.21 ± 0.72	1.82 ± 1.08	2.43 ± 1.45	1.07 ± 0.87	1.61 ± 1.31	2.15 ± 1.75	1.00 ± 0.95	1.51 ± 1.42	2.01 ± 1.90
P	0.62 ± 0.29	0.92 ± 0.44	1.23 ± 0.59	0.54 ± 0.35	0.81 ± 0.52	1.07 ± 0.69	0.50 ± 0.37	0.75 ± 0.56	1.00 ± 0.74
Mg	0.05 ± 0.03	0.07 ± 0.05	0.09 ± 0.07	0.05 ± 0.04	0.07 ± 0.06	0.09 ± 0.09	0.05 ± 0.05	0.07 ± 0.07	0.09 ± 0.10
Na	0.13 ± 0.14	0.20 ± 0.21	0.26 ± 0.27	0.15 ± 0.18	0.23 ± 0.27	0.31 ± 0.37	0.17 ± 0.21	0.25 ± 0.32	0.33 ± 0.43
All sex									
K	0.17 ± 0.15	0.26 ± 0.23	0.35 ± 0.31	0.17 ± 0.18	0.26 ± 0.27	0.35 ± 0.36	0.17 ± 0.20	0.26 ± 0.30	0.34 ± 0.40

BW, body weight; ADG, average daily gain.



mg/kg BW and 8.1% for Na, respectively (Figure 2). When the 95% confidence limits of requirements for maintenance of Ca and P were evaluated, the uncertainty of maintenance for males was greater. In addition, the uncertainty of Mg, Na, and K for maintenance were lower. The dietary mineral requirements are shown in Table 8.

4. Discussion

Information on mineral requirements for growth and maintenance is essential for herds to reach their maximum productive potential. The results of the present study emphasize the importance of updating the mineral recommendations for sheep considering the effect of sex.

As the animal grows, the body mineral content increases at similar rates early in life, decelerating later, and reaching a plateau after maturity (30, 31). One of the factors that affect this pattern of deposition mineral is sex class. Females have greater Ca and P requirements than non-castrated males. Sex may affect the mineral requirements for weight gain because females closer to maturity have higher levels of estrogen (32), which can modulate mineral dynamics in the body.

Braithwaite and Riazuddin (33) reported the effect of growth hormone on Ca metabolism, with a greater retention and an increased rate of bone Ca in ewes close to maturity. In this phase, Ca and P are mainly involved in the completion of bone development. Estradiol indirectly modulates bone Ca deposition, promoting the closure of the epiphyseal cartilage (34) due to the replacement of chondrocytes by osteogenic cells (35). Estrogen promotes osteoclast apoptosis (36) and thus decreases bone resorption, which explains

why sex affects the requirements for Ca and P, implying greater requirements for these minerals for females.

The influence of sex in the modulation of the dynamics of nutrients and the metabolic activity of the tissues due to the variation in the levels of androgenic hormones (37) can also explain the uncertainty. Irrespective of sex, the uncertainty increased as BW increased from 10 to 30 kg of BW when the 95% confidence limits were evaluated. For Ca and P, this may be associated with the dilution effect caused by the increase in body fat, since fat has a low mineral content. Body fat is the item that varies the most, while fat-free dry mass is quite constant (31) since the main change in body composition that occurs with animal growth and development is the increase in fat content and this mechanism represents the degree of maturity of the animal.

This variation of body composition explains the higher levels of uncertainty with increasing BW. In our study, sheep weighing 30 kg and 150 g/day ADG had NCag and NPg values of 1.38, 1.57, and 1.51 g/day and 0.66, 0.78, and 0.75 g/day for non-castrated males, castrated males, and females, respectively. Regardless of sex class, the estimates of NCag and NPg were similar to those suggested by CSIRO (38), of 1.10 and 0.66 g/day, respectively. However, the CSIRO (38) considers weight at maturity in estimating gain, in addition to using different methodologies and genotypes from the present study.

In our study, the NCa_m and the retention coefficient were 23.70 mg/kg BW and 54.30%, respectively. Irrespective sex, the NCa_m for animals with 30 kg BW was 710 mg Ca/day (0.710g), corresponding to a dietary requirement of 4.05 g/day considering a Ca retention coefficient of 54.30%. This dietary Ca requirement was 29% higher than that recommended by the NRC (12) but 38% lower than that

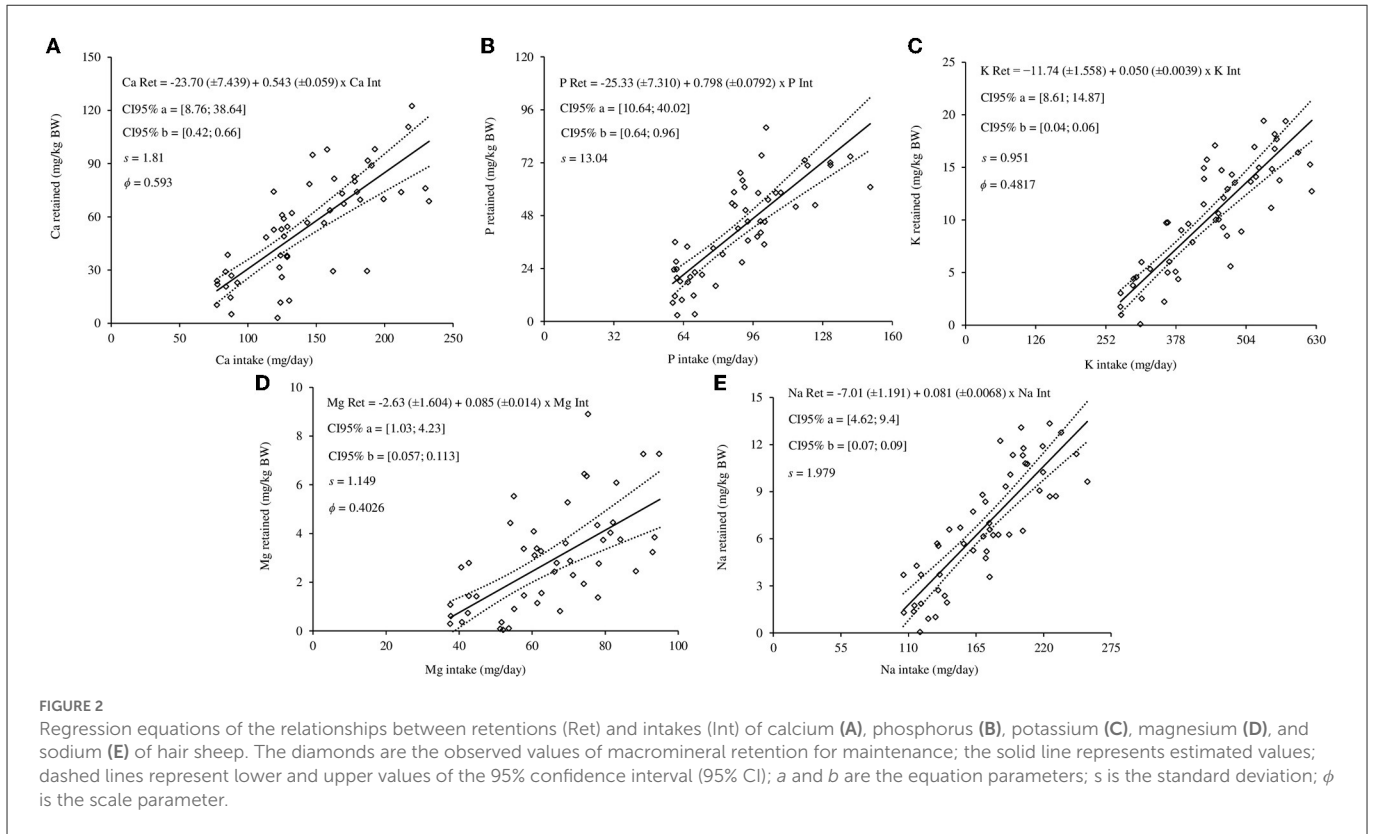


TABLE 7 Information criteria of fitted models to estimate the net requirements for maintenance of hair sheep.

Macrominerals	Variance functions	AICc	Δ	w	ER	Θ
Ca	Equation (4)	452.20	2.90	0.14	4.26	3.00
	Equation (5)	451.20	1.90	0.24	2.59	4.00
	Equation (6)	449.30	0.00	0.62	1.00	4.00
P	Equation (4)	405.20	0.00	0.44	1.00	3.00
	Equation (5)	406.30	1.10	0.25	1.73	4.00
	Equation (6)	405.90	0.70	0.31	1.42	4.00
K	Equation (4)	263.50	2.20	0.19	3.00	3.00
	Equation (5)	263.10	1.80	0.23	2.46	4.00
	Equation (6)	261.30	0.00	0.57	1.00	4.00
Mg	Equation (4)	205.10	1.80	0.22	2.46	3.00
	Equation (5)	205.00	1.70	0.23	2.34	4.00
	Equation (6)	203.30	0.00	0.55	1.00	4.00
Na	Equation (4)	170.80	0.00	0.62	1.00	3.00
	Equation (5)	173.20	2.40	0.19	3.32	4.00
	Equation (6)	173.20	2.40	0.19	3.32	4.00

AICc, Akaike information criterion corrected for small samples; Δ , Akaike difference; w, model probability; ER, evidence ratio computed for each model; Θ , number of parameters of the linear model.

recommended by the INRA (39). The recommended NP_m in our study was 25.33 mg/kg BW and the retention coefficient was 80%. Irrespective of sex, hair sheep had a P dietary requirement of 1.87 g/day. This value is close to the NRC value (12), which is 1.98 g/day for sheep.

The estimation of macromineral requirements for sheep varies between the different Committees, being the main factors for

variation, the differences in the values adopted for maintenance requirements and the coefficient of absorption or retention of minerals. The contrasting results of these coefficients are due to endogenous losses that are associated with variations in voluntary intake that directly influence maintenance requirements.

The use of the true retention coefficient seems to be more accurate than the absorption coefficient when it comes to estimating

TABLE 8 Dietary mineral requirements for hair sheep.

BW (kg)	ADG (g/day)	Non-castrated males				Castrated males				Female				All sex
		Dietary minerals requirements (g/day)												
		Ca	P	Mg	Na	Ca	P	Mg	Na	Ca	P	Mg	Na	
10	100	2.54	1.03	0.77	2.25	2.68	1.09	0.81	2.32	2.67	1.09	0.92	2.48	5.83
	150	3.59	1.39	0.98	2.95	3.81	1.48	1.05	3.05	3.79	1.48	1.20	3.29	7.58
	200	4.64	1.75	1.20	3.64	4.93	1.86	1.29	3.78	4.91	1.86	1.47	4.09	9.32
20	100	2.70	1.24	1.06	2.94	2.91	1.33	1.16	3.23	2.85	1.31	1.25	3.61	8.15
	150	3.61	1.54	1.27	3.55	3.92	1.67	1.41	3.97	3.84	1.65	1.53	4.56	9.88
	200	4.53	1.84	1.47	4.15	4.94	2.02	1.66	4.72	4.83	1.98	1.80	5.50	11.61
30	100	3.01	1.50	1.38	3.72	3.24	1.60	1.50	4.11	3.16	1.58	1.58	4.64	10.48
	150	3.85	1.78	1.57	4.29	4.20	1.93	1.76	4.87	4.09	1.89	1.86	5.66	12.20
	200	4.70	2.05	1.77	4.85	5.16	2.25	2.01	5.63	5.01	2.20	2.12	6.69	13.92

BW, body weight; ADG, average daily gain.

mineral requirements, as the minerals being absorbed will not always be used by the animal. It is important to emphasize that mineral requirements must be considered, as the exact demand for mineral needs can reduce mineral excretion and environmental pollution, especially that caused by excessive phosphorus excretion. The imbalance in P levels contributes to soil contamination, which when carried by rain can contribute to water eutrophication (40). The proposal of the equations for P aims to meet the requirements of growing animals, minimizing the use of P in the diet because high dietary concentrations may increase fecal and urinary excretion of the mineral.

Differences in the requirements for Mg were observed for animals during the evaluated growth phase. The relationship between Mg and energy metabolism may explain the effects of sex on the requirements of female hair sheep. Commonly, females initiate fat deposition more quickly when compared to males (12). Thus, the composition of the gain could explain the higher Mg requirement in females, compared to males, as they deposit greater amounts of fat (41).

Considering the role of Mg in the cell's energy metabolism, it is likely that the Mg requirements of females are in line with the greater energy requirements for maintaining females at the peak of growth, coinciding with sexual maturity. In addition, in the closing of the bone epiphysis, a greater supply of vitamin D is required because various stages of vitamin D conversion are actively dependent on the bioavailability of Mg (42, 43), contributing to maintaining the physiologic functions of the musculoskeletal system.

Magnesium, along with K and Na, is a cation extensively distributed in the body, around 70% is associated with the skeleton, 25% with the muscle mass, and only around 1% within the extracellular space (44). Since Mg metabolism is not considered to be under hormonal control, the variation of body composition and, consequently of Mg in the bone and tissues can explain the uncertainty as the animal grows and approaches maturity.

In the current study, sex did not affect K and Na requirements for maintenance. The Na and K are required for nutrient absorption processes in the gastrointestinal tract, and for maintenance of the acid-base balance in the animal's body (9). All these processes are

essential for life. Thus, due to the role in homeostatic control, the requirement for maintenance did not vary between sex. In our study, the requirements for K and Na were 0.35 and 0.21 g/day, respectively, for the maintenance of hair sheep with 30 kg of BW. These values are lower than those recommended by the NRC (12) and INRA (39) that suggests 4.10 and 3.15 g/day for K and 0.32 and 0.45 g/day for Na, respectively. Thus, our data indicate that both Committees overestimate K and Na requirements for the maintenance of hair sheep.

Sex did not affect K requirements for growth. For animals weighing 30 kg BW with 150 g ADG (136 g EBWG), the NK_g correspond to 0.258 g/day. In this context, the value suggested by our study is close to the recommendations of the NRC (12) and INRA (39), which corresponds to 0.270 g/day. However, the sex influences NNa_g , and females demand greater amounts of Na with increasing BW compared to males. Regarding the ARC (7) and Suttle (9), the Na recommendation for sheep is around 1.10 g/kg of weight gain, a value also suggested by the NRC (12). The body concentration of Na is about 1.3 g/kg BW (38), of which about a third is in the bones and most of the remainder in the extracellular fluids (2). Sodium present in bones is adsorbed on the surfaces of hydroxyapatite crystals within long bones and therefore does not interact with other ions present in body fluids (45).

The Na requirements are related to metabolic changes that occur as the animal grows. There is an inverse relationship between water and fat in the animal's body; with increasing fat content, the water content decreases (3). The increase in body fat deposition in females close to mature weight can alter the body Na balance. This occurs because the reduction in water content increases the concentration of Na in the extracellular fluid, which increases the glomerular filtration rate and reduces the efficiency of tubular absorption. These metabolic changes increase the Na excretion rates and may therefore be associated with a higher Na requirement as the BW increases (45).

In conclusion, our study brings a significant contribution since it shows the influence of sex on mineral requirements of hair sheep breed of Santa Ines, Morada Nova, Brazilian Somali and crossbreed. The confidence limits show that the models proposed to estimate the

nutritional requirements of macrominerals for growing hair sheep were viable. This represents an advance in the understanding of nutritional requirements because the confidence limits in mineral requirements has not been reported by the committees, and requirements are considered as mean values. The recommendations for confined sheep of different sexes in the growth phase of 10 to 30 kg BW allow the nutritionist to recommend the mineral requirements more reliably, which implies a reduction in costs and environmental impact. The information generated in this study can be used to optimize the mineral management of hair sheep in the growing phase in warm areas.

Data availability statement

The original contributions presented in the study are included in the article/[Supplementary material](#), further inquiries can be directed to the corresponding author.

Ethics statement

The approval of the ethics committee on animal use was not necessary for this study because the data were collected from previously published sources.

Author contributions

CH, AB, and MME: data curation and investigation. MA, LS, and MMA: methodology, formal analysis, and validation. PM, LC, LB, and RO: methodology and validation. EP: conceptualization, methodology, writing—original draft, writing—review and editing, and project administration. All authors made important

contributions to this manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fvets.2023.1032429/full#supplementary-material>

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