









Detoxified castor in the diets of dairy goats: I. Effects on intake, digestibility, and renal and hepatic parameters

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ABSTRACT - The objective was to evaluate the intake, digestibility, nitrogen balance, and metabolic profile of lactating goats fed diets containing detoxified castor cake (DCC) by alkaline solutions during 150 days of lactation. Twenty-four Saanen and Anglo Nubian goats, approximately 17 months old (first lactation) and body weights of 43±2.97 kg, were distributed in a completely randomized block design with eight replicates. Treatments consisted of three diets, one containing soybean meal (SM) and two others containing DCC, with calcium hydroxide [Ca(OH)₂] and sodium hydroxide (NaOH). The diets significantly influenced the intake of dry matter (DM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), and total digestible nutrients (TDN). We observed a higher dry matter intake (DMI) in goats fed SM, similar to that of goats fed Ca(OH)₂ DCC. Intake of nutrients followed the same trend as DMI. There was no significant effect of diets on digestibility of DM, CP, EE, and NDF; however, we observed a significant effect of diets on the levels of nitrogen intake (NI) and urinary nitrogen (UN). The goats fed SM consumed a larger quantity of nitrogen, but all had the same nitrogen balance, indicating that goats fed DCC were more efficient. The diets did not influence renal and hepatic parameters. Inclusion of castor cake in the diet of goats in confinement is an attractive option, considering that goats fed DCC present lower feed conversion, and its use does not cause hepatic and renal alterations, suggesting that SM can be completely replaced.

Keywords: Anglo Nubian, persistence, ricin, Saanen, toxicity

Introduction

The recent increase in the inclusion of biodiesel in the world energy matrix has led to the production of ruminant feeds from byproducts or cakes obtained after extraction of oil from oilseeds, which constitute the main byproducts of the biodiesel production chain. Thus, a possibility of integrating the agroenergy and agricultural chains and generating employment and income has emerged, in addition to possibly minimizing the environmental problems caused by these residues.

In this context, studies that promote generation of information regarding best practices for the use of byproducts in ruminant feeding are necessary. Among the byproducts generated from the biodiesel chain, castor bran and castor cake are important, since the cultivation of castor seed is increasing

every year mainly due to its low demand for water and soil nutrients. One of the main advantages of using castor (*Ricinus communis*) byproducts in ruminant feeding is its high protein value. Soybean meal (SM) is a feedstuff primarily intended for human consumption, thereby increasing its production costs (Oliveira et al., 2010a; Borja et al., 2017). According to Akande et al. (2015), castor cake contains from 342 to 480 g of crude protein (CP) per kilogram of dry matter; however, after the detoxification process, this CP content may decrease depending on the alkaline product used; the true protein may be insoluble in neutral detergent (Oliveira et al., 2010b).

Furthermore, castor bean byproducts present a nutritional limitation due to the presence of toxic proteins such as ricin and ricinus agglutinin, besides ricinin alkaloids and allergenic complexes, which, when ingested by animals, trigger inactivation of ribosomes, hemolysis, diarrhea, and allergic attacks (Dang and Vam Damme, 2015). Nevertheless, to circumvent this situation after the detoxification process, castor cake can be a viable alternative for use as animal feed (Anandan et al., 2005), considering the possibility of using animal byproducts from the biodiesel chain in diets for ruminants, giving efficient allocation to these products, and incorporating them in the productive chain of dairy goats.

Thus, the hypothesis of this study is that there is a possibility of replacing soybean meal for castor cake detoxified by alkaline solutions at the lactation stage of Saanen and Anglo Nubian goats without adverse effects on digestibility and metabolic parameters. Based on the above, we aimed to evaluate the influence of castor detoxified by alkaline solutions on the intake, digestibility, nitrogen balance, and renal and hepatic metabolic profiles of lactating goats.

Material and Methods

All animal procedures were conducted in accordance with the regulations of the local Ethics Committee on the Use of Animals (case no. 005/2015). The chemical analyses were performed in Sobral, Ceará, Brazil (3°44'57.42" S, 40°20'43.50" W). The experiment with goats was conducted in Sobral.

Twenty-four goats with 43±2.97 kg body weight and body condition scores of 2.5±0.5 were used. Treatments consisted of three diets: the first formulated with corn and soybean meal (SM) and the other two with detoxified castor cake (DCC) using calcium hydroxide (Ca(OH)₂ DCC) or sodium hydroxide (NaOH DCC), as a total substitution of SM. Tifton 85 hay, chopped to 4 cm, was used as a forage source.

Goats were distributed in a randomized block design (breed factor), with eight replicates per diet. At the end of the survey, there was a preliminary analysis of the data, in which we evaluated a possible interaction between breeds and diets, which that did not happen. Therefore, we chose to use the randomized blocks to assess the effect of the breeds. In this way, all the remaining variables had eight replicates, because the effect of breeds was null.

The goats were confined and housed in individual masonry stalls with suspended wooden floors and a total area of 5.06 m². Each stall contained a 2.87-m² solarium, constructed with wooden slats, which assured visual, auditory, olfactory, and tactile contact with the other animals of the adjacent bays. The solarium area was composed of wooden grids equipped with feeders, drinkers, and saltshakers. During the pre-experimental period, the goats were identified, treated against ecto- and endoparasites, and vaccinated against rabies (Dectomax[®] and Ourovac[®]). Then, they were distributed among treatments, and a 15-day adaptation period to the diets was allowed.

The experimental diets were formulated based on the isonitrogenous and isoenergetic diet recommendations of the NRC (2007) for goats with 45 kg body weight and daily milk production of 1.5 L. The chemical compositions of the ingredients used in the diet preparation are described in Table 1, and the proportions of ingredients and chemical composition of the diets are shown in Table 2. Detoxification with calcium hydroxide significantly elevated the calcium levels in the Ca(OH)₂ DCC diet. Upon addition of 90 g Ca(OH)₂, each kilogram of castor meal received 22.25 g of calcium (Table 1).

This addition represents 40% of the calcium present in the mineral supplement and could cause an imbalance in the dietary calcium:phosphorus ratio. Owing to this variation, the forage:concentrate ratio differed across the treatments. Moreover, DCC should be included as a protein ingredient at a maximum proportion of 8% of dietary dry matter (Pompeu et al., 2012).

Table 1 - Chemical composition of the ingredients used in the experimental diets

Item (g/kg dry matter)	Ingredient				
	Tifton 85 hay	Ground corn	Soybean meal	Ca(OH) ₂ DCC ¹	NaOH DCC ²
Dry matter (g/kg fresh matter)	872.50	889.20	870.20	904.20	904.80
Organic matter	911.30	965.90	956.90	867.70	855.60
Mineral matter	88.70	34.10	43.10	132.30	144.40
Crude protein	104.10	79.50	443.30	315.40	309.00
Neutral detergent insoluble protein	26.98	30.23	131.75	100.27	102.74
Acid detergent insoluble nitrogen	12.26	20.92	40.03	48.79	49.35
Ether extract	14.50	36.80	28.80	52.10	47.50
Total carbohydrates	792.80	845.70	484.70	500.10	492.60
Non-fiber carbohydrates	277.80	722.40	320.80	103.90	132.40
Neutral detergent fiber (NDF)	722.70	184.60	217.80	483.40	443.50
NDF corrected for ash and protein	514.90	123.20	163.80	396.10	360.10
Acid detergent fiber	472.20	69.00	117.90	379.20	388.70
Lignin	60.60	8.80	12.20	50.70	46.10
Total digestible nutrients	546.80	848.00	822.50	620.50	627.90

¹ Ca(OH)₂ castor cake: 0.9 g Na/kg DM and 22.25 g Ca/kg DM.

² NaOH castor cake: 29.2 g Na/kg DM and 0.63 g Ca/kg DM.

Table 2 - Ingredient proportions and chemical compositions of the experimental diets

Item	Diet		
	Soybean meal	Ca(OH) ₂ DCC	NaOH DCC
Ingredient (g/kg dry matter)			
Tifton 85 hay	525.40	485.80	474.30
Ground corn	414.20	424.60	437.40
Soybean meal	58.70	-	-
Detoxified castor cake (DCC)	-	89.60	85.70
Limestone	1.70	-	2.6
Mineral mixture ¹	<i>ad libitum</i>	<i>ad libitum</i>	<i>ad libitum</i>
Chemical composition (g/kg dry matter)			
Dry matter (g/kg fresh matter)	883.03	890.84	885.76
Organic matter	939.17	933.06	930.90
Mineral matter	62.49	66.94	71.43
Crude protein	113.94	110.13	112.12
Neutral detergent insoluble protein	12.54	13.58	13.89
Acid detergent insoluble nitrogen	3.17	3.65	3.54
Ether extract	26.46	29.22	29.82
Total carbohydrates	759.80	766.30	756.60
Non-fiber carbohydrates	471.80	468.70	477.40
Neutral detergent fiber (NDF)	416.78	424.98	404.54
NDF corrected for ash and protein	287.97	297.65	279.21
Acid detergent fiber	352.80	356.60	337.19
Lignin	30.86	32.62	30.32
Total digestible nutrients	674.90	678.80	678.70

¹ Guaranteed level (per kg, inactive elements): calcium, 218 g; phosphorus, 71 g; sulfur, 20 g; manganese, 1,300 mg; potassium, 28.20 mg; cobalt, 30 mg; selenium, 15.30 mg; zinc, 1700 mg; copper, 710 mg.

Diets were supplied daily at 07.30 and 14.30 h, allowing a 10% surplus supply. Samples were collected from the bulk, concentrate, and also leftovers during the entire experimental period, then duly packaged in identified plastic bags and stored in a freezer at -8°C .

Apparent digestibility coefficients were estimated indirectly using the internal iADF indicator. To this end, feces were collected directly from the rectal bulb for 5 d at different times (0, 3, 6, and 9 h after the first feeding), aiming for greater daily representativeness. Next, they were identified and stored in a freezer at -8°C . At the end of data collection, composite samples were prepared and then dried in a forced ventilation oven at 55°C until constant weight. Fecal and food samples were incubated *in situ* for a period of 240 h, according to the methodology described by Casali et al. (2008).

For nitrogen balance evaluation, total urine production was estimated by the creatinine concentration in the urine. At the end of the growth phase, spot urine samples were obtained approximately 4 h after feeding, from spontaneous urination in colostomy bags (Medsonda[®]) with a capacity of 200 mL. Samples were prepared according to the methodology of Valadares et al. (1999) and immediately frozen. Urine production was estimated by the equation proposed by Fonseca et al. (2006).

Fecal samples were collected directly from the rectal bulb for 5 d at different times (0, 3, 6, and 9 h after the first feeding) for a representative sampling. Furthermore, the fecal samples used for digestibility tests were collected on different days; thus, two samplings were performed.

Blood samples were collected using 9.0-mL vacutainer tubes (Greiner Bio-One, Vacuette[®] Americana, SP, BRA), by puncturing the jugular vein 5 d before the end of the rearing phase and 4 h after the morning feed for the analysis of total urea and protein content in the serum.

Two blood samples were collected from each animal, one in a tube containing an anticoagulant (EDTA) and another in a tube without the anticoagulant. The samples containing anticoagulant were used for the analysis of urea and total protein dosage, while samples without the anticoagulant were used for the analysis of creatinine, total and direct bilirubin, albumin, alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyltransferase (GGT) levels. To determine urea and total protein dosage, a serum was obtained by centrifuging the tubes at $3,293 \times g$ for 15 min; then, they were identified and stored in Eppendorf[®] mini-tubes, and frozen for analysis. Blood parameters and urine creatinine were analyzed with commercial Labtest[®] kits using colorimetric procedures.

Castor cakes used in this study were obtained after collecting oil, by mechanically pressing castor bean seeds at temperatures between 90 and 100°C . After mixing the cakes with reagents and water for 3 h (mixing for 10 min and resting for 30 min, alternately), the cakes were placed outdoors on a plastic canvas for 48 h and constantly rolled with a squeegee adapted for homogeneous drying. After drying, the cakes were chopped using a forage machine to reduce the material size and to facilitate its homogenization with the other ingredients.

The concentrations of alkaline products (calcium hydroxide and sodium hydroxide) used for 100% detoxification of ricin in crude castor cakes were 90 g $\text{Ca}(\text{OH})_2$ and 60 g NaOH per kilogram, respectively, which were diluted in 2 L of water using a stationary mixer (Fischer[®] MOB 400 G2) equipped with a three-phase motor. No hemagglutinating activity was observed at those concentrations, i.e., ricinus agglutinin was no longer active; therefore, these two concentrations were used to formulate the diets.

The concentrations of NDF were corrected for ash and nitrogen as proposed by Mertens et al. (2002), and for proteins according to Licitra et al. (1996). The acid detergent lignin (ADL) fraction was extracted with 72% sulfuric acid (Van Soest et al., 1991). Non-fibrous carbohydrate (NFC) content was calculated by an adaptation of the method proposed by Hall (2003), using NDF corrected for ashes and protein (NDFap). Total carbohydrate (TC) content was obtained using the equation proposed by Sniffen et al. (1992).

The quantity of total digestible nutrients (TDN) was calculated according to Weiss (1999). The TDN values were converted into net energy (NE) for production and digestible energy (DE), according to

the equations suggested by the NRC (2001). The TDN intake (TDNI) was calculated according to the methodology described by Sniffen et al. (1992), as follows:

$$\text{TDNI} = (\text{CPI} - \text{CPf}) + 2.25(\text{EEI} - \text{EEf}) + (\text{TCI} - \text{TCf}),$$

in which CPI, EEI, and TCI correspond to the intakes of CP, EE, and TC, respectively; and CPf, EEf, and TCf represent the respective excretion of CP, EE, and TC in the feces.

The levels of nitrogen intake (NI), fecal nitrogen (FN), urinary nitrogen (UN), and nitrogen excreted in the milk (NM) were determined by the micro Kjeldahl method (no. 954.01) of the AOAC (2003). The retained nitrogen (RN) was calculated according to the equation: $\text{RN} = \text{NI} - (\text{FN} + \text{UN} + \text{NM})$. Before each milking, during the entire experimental period, the teats of the goats were sanitized with sodium pre-dipping and dried with paper towels, and at the end of milking, another cleaning solution was applied with post-dipping for protection and prevention against mastitis.

Data were initially subjected to normality (Shapiro-Wilk) and homoscedasticity (Levene) tests and to analysis of variance by the F test when the presuppositions were met, using the following model:

$$Y_{ij} = \mu + \alpha_i + \beta_j + \varepsilon_{ij},$$

in which Y_{ij} is the dependent variable corresponding to the experimental observation, μ is the overall mean, α_i is the fixed effect of the diets, β_j is the fixed effect of the breed, and ε_{ij} is the random error, assuming an independent normal distribution. A comparison of means was performed by Tukey's test at 5% probability to evaluate the effects of breed and diet. Statistical analysis was performed using the PROC MIXED procedure of SAS software (Statistical Analysis System, version 9.4).

Results

The diets significantly influenced ($P < 0.05$) the intake of DM and other nutrients; however, no significant ($P > 0.05$) effect of the different breeds was observed on intake (Table 3). We observed a higher dry matter intake (DMI) for the goats fed SM (2318.10 g/day) and Ca(OH)_2 DCC (2220.28 g/day), and lower DMI for the goats fed NaOH DCC (1961.38 g/day). The intake of CP, EE, NDF, and TDN exhibited the same behavior observed for the DMI, in which the goats fed the SM and NaOH DCC diets ingested larger quantities, except for DMI based on body weight (BW), in which the DMI %BW of goats fed Ca(OH)_2 DCC did not differ from that of goats fed NaOH DCC.

There was no significant effect ($P > 0.05$) of diets or of breeds on the digestibility of DM and nutrients during the lactation phase of goats (Table 4). We observed a significant effect ($P < 0.05$) of diets on the levels of nitrogen intake (NI) and urinary nitrogen (UN), but no significant effect was observed ($P > 0.05$) for the breeds on the other parameters of nitrogen balance (Table 5). Goats fed SM exhibited higher NI (55.79 g/day), followed by goats fed DCC. Similarly, UN of goats fed SM (6.95 g/day) was higher but did not differ from that of goats fed the Ca(OH)_2 DCC (5.94 g/day) or NaOH DCC (5.88 g/day) diets.

Table 3 - Intake of dry matter and nutrients of goats fed diets containing detoxified castor by different alkali in substitution to soybean meal

Item	Diet			SEM	P-value	
	Soybean meal	Ca(OH)_2 DCC	NaOH DCC		Diet	Breed
Dry matter	2318.10a	2220.28a	1961.38b	44.10	<0.05	0.39
Dry matter (% of body weight)	4.53a	4.17ab	3.99b	0.106	<0.05	0.86
Crude protein	259.62a	255.50a	219.67b	4.976	<0.05	0.39
Ether extract	122.82a	120.01a	101.99b	2.332	<0.05	0.76
Neutral detergent fiber	1398.86a	1334.35a	1216.05b	28.15	<0.05	0.33
Total digestible nutrients	1460.40a	1443.18a	1294.51b	21.17	<0.05	0.21

DCC - detoxified castor cake; SEM - standard error of the mean.

Means within a row with different letters are different by Tukey's test at 5% significance.

For hepatic and renal parameters, it was observed that there was no significant effect ($P>0.05$) of diets or breeds on the blood levels assessed (Table 6).

Discussion

The goats fed NaOH DCC presented lower DMI, approximately 356 g DM/day less than goats that received a standard diet based on SM. The reduction in the DMI can be associated to the presence of sodium in the diet, which serves as an intake controller (Yousfi et al., 2016). The amount of sodium in the NaOH DCC was 32.4 times higher than that in the Ca(OH)_2 DCC diet (Table 1), which further highlights its effect in the control of DMI. Araújo et al. (2018) evaluated the replacement of SM by NaOH DCC in the diet of goat kids during the growth phase and also observed a reduction in DMI.

Table 4 - Apparent digestibility of lactating goats fed diets containing detoxified castor by different alkali in substitution to soybean meal

Digestibility (g/kg DM)	Diet			SEM	P-value	
	Soybean meal	Ca(OH)_2 DCC	NaOH DCC		Diet	Breed
Dry matter (g/kg FM)	702.33	690.29	675.69	18.69	0.61	0.91
Crude protein	714.20	702.26	687.62	18.67	0.61	0.90
Ether extract	752.28	761.46	742.01	16.76	0.71	0.87
Neutral detergent fiber	737.28	746.46	727.01	16.80	0.72	0.99

DM - dry matter; FM - fresh matter; DCC - detoxified castor cake; SEM - standard error of the mean. Means within a row with different letters are different by Tukey's test at 5% significance.

Table 5 - Nitrogen balance of lactating goats fed diets containing detoxified castor by different alkali in substitution to soybean meal

Item (g/day)	Diet			SEM	P-value	
	Soybean meal	Ca(OH)_2 DCC	NaOH DCC		Diet	Breed
Nitrogen intake	55.79a	52.12b	51.87b	1.173	<0.05	0.78
Fecal nitrogen	15.69	15.43	14.10	0.608	0.17	0.08
Urinary nitrogen	6.95a	5.94ab	5.88b	0.290	<0.05	0.05
Nitrogen in milk	7.88	7.36	7.32	0.165	0.05	0.78
Retained nitrogen	25.26	23.39	24.55	1.105	0.49	0.05

DCC - detoxified castor cake; SEM - standard error of the mean. Means within a row with different letters are different by Tukey's test at 5% significance.

Table 6 - Blood parameters of lactating goats fed diets containing detoxified castor by different alkali in substitution to soybean meal

Parameter	Diet			SEM	P-value	
	Soybean meal	Ca(OH)_2 DCC	NaOH DCC		Diet	Breed
Total proteins (g/dL)	8.93	10.84	10.50	0.69	0.15	0.42
Albumin (g/dL)	4.34	5.42	5.00	0.32	0.09	0.51
Urea (mg/dL)	40.67	39.29	36.60	1.30	0.11	0.54
Creatinine (mg/dL)	1.81	1.74	1.97	0.17	0.62	0.68
Direct bilirubin (mg/dL)	1.18	1.29	1.22	0.20	0.91	0.65
Alkaline phosphatase (IU/L)	11.18	10.61	11.15	0.93	0.88	0.63
Alanine aminotransferase (IU/L)	102.63	76.22	92.55	11.45	0.42	0.90
Aspartate aminotransferase (IU/L)	19.18	15.06	15.06	2.48	0.28	0.82
Gamma-glutamyltransferase (IU/L)	64.19	77.73	64.40	4.56	0.08	0.36

DCC - detoxified castor cake; SEM - standard error of the mean.

The mean value observed for DMI was found to be close to 4%. Animals of this size and category can consume up to 4.5% of their BW per day (Teixeira et al., 2011); however, these values are not well defined, given the differences in the feeding behavior and processing capacity of diets in the gastrointestinal tract of goats (Forbes, 2007).

The CP intake changed with the replacement of SM by the NaOH DCC diet. The effect on DM intake influenced CP intake results, which have a direct relationship since the diets were isonitrogenous. According to the NRC (2007), CP intake of goats with a BW of 45 kg and daily production of 1.5 L of milk is 228 g/day. The CP intake observed in this experiment, therefore, met the requirements of goats for this nutrient. Although the goats fed the NaOH DCC diet had a lower CP intake in relation to the recommended by the NRC (2007), milk production was 30.55% higher, demonstrating greater efficiency in the use of this nutrient by these animals, considering that the amount of RN was equal for all treatments (Table 5).

The RN was similar for the three diets evaluated, with values close to 50%, indicating efficiency in the use of this nutrient (Rapetti et al., 2014). Notably, there is a positive relationship between RN and milk production (Lapierre et al., 2005), since the positive balance means that nitrogen retention was sufficient to meet the requirements of metabolizable protein, besides being a good estimate of the quality of nitrogen available to form body tissues, including those of the mammary glands (Safayi and Nielsen, 2013). Excess nitrogen, however, can be detrimental to body metabolism (Alonso-Mélendez et al., 2016). Thus, higher total protein and urea levels were expected to be found in the blood circulation of these animals (Table 6); however, this was not the case, which indicates that there was no excess of ammonia in the rumen, since the excretion of urea was also equal.

Notably, urea recycling provides greater energy expenditure, as the formation of 1 mol of urea uses 2 mol of ATP, and for which there is efficient recycling and excretion of urea, requiring greater efficiency of the liver and kidneys, respectively (Owens and Zinn, 1988). This could increase the levels of creatinine, direct bilirubin, ALP, ALT, AST, and GGT (Table 6); however, despite the absence of the effect of diets or breeds on the blood parameters examined, concentrations of the analyzed variables remained within the normal reference values (Kaneko et al., 2008). For the animals in which the concentrations of creatinine, direct bilirubin, total protein, albumin, and urea were within the reference interval, we did not find higher levels of milk production than in animals in which these concentrations were reduced (Contreras et al., 2000).

When protein intake does not meet the nutritional requirements of animals in lactation, the demand for amino acids for protein synthesis in milk reduces the synthesis of other proteins, especially albumin (Contreras et al., 2000); as the levels were normal, this suggests that the protein intake requirements were met. In contrast, the enzymes ALT, AST, and GGT are also good indicators of liver injury in ruminants, provided that the possibility of muscle and cardiac lesions are also considered, and it is associated with the occurrence of ricin poisoning in sheep (Aslani et al., 2007). The DCC, independent of the alkaline product for detoxification, therefore, did not cause liver problems in goats.

The effect of the DMI influenced the intake of EE, NDF, and TDN, which maintain a direct relationship, mainly because the diets were isonitrogenous (Table 2), and selectivity was crucial to the intake of these components of the diet. In relation to TDN intake, the highest values for goats fed SM is explained by the greater amount of EE consumed (Danieli and Ronchi, 2018). According to the NRC (2007), the recommended TDN intake for goats in this category is 1,200 g TDN/day, which indicates that all diets met TDN requirements, even the NaOH DCC diet (1294 g TDN/day).

Conclusions

The results of the present study suggest that the replacement of soybean meal with detoxified castor with alkaline solutions is a viable alternative for feeding lactating goats. Diets formulated with detoxified castor bean cake controls intake, but do not adversely affect liver and kidney function and nitrogen balance.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Conceptualization: R.A. Araújo. Data curation: R.A. Araújo. Formal analysis: R.A. Araújo and R.C.F.F. Pompeu. Funding acquisition: R.A. Araújo and R.C.F.F. Pompeu. Investigation: R.A. Araújo, R.C.F.F. Pompeu, M.C.P. Rogério, S.R. Maranhão and J.N.M. Neiva. Methodology: R.A. Araújo, R.C.F.F. Pompeu, M.J.D. Cândido, M.C.P. Rogério, R.C. Lucas, S.R. Maranhão, C.F. Santos Neto and J.N.M. Neiva. Project administration: R.A. Araújo, R.C.F.F. Pompeu and C.F. Santos Neto.

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