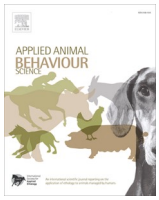
Applied Animal Behaviour Science 278 (2024) 106362

Contents lists available at ScienceDirect

Applied Animal Behaviour Science

journal homepage: www.elsevier.com/locate/applanim



Ewe-lamb bond at birth and during lactation in an equatorial semi-arid environment is better in a native than in an introduced breed

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ARTICLE INFO

Keywords:
Lambing
Lactation
Maternal care
Suckling
Sheep

ABSTRACT

Santa Ines and Dorper sheep are the most popular breeds in the Brazilian semi-arid region. The former is of local origin and less intensively selected for productivity, and the latter is an introduced exotic breed selected for meat production. Dorper lambs are heavier at birth than Santa Ines lambs, which might increase the need for birth assistance, negatively influencing the newborn and maternal behaviours. Dorper lambs grow faster, most likely influencing their behavioural strategies during lactation, depending less on their mother than Santa Ines lambs. The aim of this study was to compare the behaviours of Santa Ines and Dorper ewes and their lambs at birth and throughout lactation. A complementary aim was to determine if being single or twin lambs impact differently according to the breed. Twenty-five Santa Ines ewes and their 32 lambs (18 single and 14 twins) and 21 Dorper ewes and their 26 lambs (16 single and 10 twins) were used in the study. Mother-offspring behaviours were recorded from birth until 63 days after lambing. Dorper ewes had a greater incidence of dystocia ($P = 0.02$) and longer labour ($P = 0.0001$) than Santa Ines ewes. At birth, Santa Ines ewes displayed a greater frequency of low-pitched bleats ($P = 0.0006$) and tended to spend more time grooming their lambs than Dorper ewes ($P = 0.07$). Immediately after birth, Santa Ines lambs shook their head earlier than Dorper lambs ($P = 0.003$). The litter size effects were unrelated to the breed: ewes of single deliveries groomed their lambs earlier ($P = 0.04$), and those that delivered twins spent more time grooming the first lamb delivered ($P = 0.01$). Over the first two weeks after parturition, Santa Ines ewes displayed higher frequencies of head-up postures ($P = 0.0001$), high-pitched bleats ($P = 0.02$) and smelled their lambs more times ($P = 0.0001$) than Dorper ewes. Santa Ines lambs were closer to their mother when grazing ($P = 0.0001$), while Dorper lambs were closer to their mother when lying down ($P = 0.049$). In conclusion, Santa Ines sheep shows a stronger ewe-lamb bond than Dorper soon after parturition, and such differences persist at least until 9 weeks after lambing. The ewe-lamb bond was weaker in twin than single lambs, without differences in the degree of its strength between Santa Ines and Dorper sheep.

1. Introduction

A total of 0.4 billion sheep worldwide are reared in semi-arid regions (dos Santos et al., 2023). In particular, the largest sheep flock in Brazil is composed of hair breeds raised under extensive and small-scale farming systems in semi-arid conditions (de Azambuja Ribeiro and González-García, 2016), where the mortality rate of lambs is up to 30 % (dos Santos et al., 2023). Santa Ines is the native breed of hair sheep with the largest population in Brazil (Costa et al., 2012), well adapted to the said production systems, with a good maternal ability (de Azambuja Ribeiro and González-García, 2016). However, the growth performance of their lambs is poorer than that of breeds specially developed for meat

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https://doi.org/10.1016/j.applanim.2024.106362
Received 10 January 2024; Received in revised form 16 July 2024; Accepted 25 July 2024
Available online 26 July 2024

Table 1
Mother-offspring behaviours at birth.

Behaviour	Definition	Unit
Ewes		
Latency to groom	The time between lamb birth and the first time at which the ewe licked any part of the body of her lamb	Duration (s)
Low pitched bleats	Ewe vocalized with the mouth closed	Frequency
Grooming	Ewe licked/cleaned the newborn lamb	Duration (min)
Lambs		
Low pitched bleats	Vocalization with the mouth closed	Frequency
Seeking udder	Lamb stayed in the parallel inverse position with its head nudging the ewe in regions close to the udder	Duration (min)
Latency to head shaking	The time between lamb birth and the first time the lamb raised and shook its head	Duration (s)
Latency to stand up	The time elapsed from birth until the lamb put its four legs on the ground over at least five seconds	Duration (min)
Latency to suckle	Time elapsed from birth until the newborn performed its first successful suckling over at least five seconds	Duration (min)

Table 2
Description of the behavioural categories of ewes and lambs throughout the lactation.

Behaviour	Description	Unit
Ewes		
Grazing	Ewe standing or walking along the paddock with their head directed to the ground, biting, chewing, pulling and searching for grass	*Duration (min)
Lying down	Ewes with flank in physical contact with the ground surface	*Duration (min)
High-pitched bleats	Ewe vocalized with open mouth	Frequency
Low pitched bleats	Vocalization with the mouth closed	
Head-up posture	Ewe standing inactive, head raised above the body, ears alert and forwards, while vocalizing with open mouth	Frequency
Nose contact	Ewe touches any body part of the lamb with her nose	Frequency
Lambs		
Grazing	Lamb standing or walking along the paddock with head down, biting, chewing or searching for grass	*Duration (min)
Lying down	Lamb with flank in physical contact with the ground	*Duration (min)
Close to the mother	Lamb was apart less than two times the ewe's body length from its mother	*Duration (min)
Within the creep-feeding	Lamb standing, lying down or eating inside the creep-feeding	*Duration (min)
successful suckling	Lamb gain access to the udder and suckle for more than 5 s	Frequency
Suckling attempt	Lamb gain access to the udder and suckle for more than 5 s (Successful attempt) or less than 5 s (Unsuccessful attempt)	Frequency
High-pitched bleats	Lamb vocalizes with open mouth	Frequency
Head-up	Lamb standing with the heat raised above the body, ears, alert, while vocalizing with open mouth	Frequency

* Variables expressed as a proportion of time. The proportion of time was calculated by dividing the time spent by the ewes/lambs in one activity by the total time of observation.

production (Cartaxo et al., 2011; Tsegay et al., 2013). Dorper is another hair breed widely reared in Brazil, originated in South Africa, also well adapted to semi-arid environments and selected for meat production. This breed was introduced in Brazil to enhance lamb growth, shortening the time from birth to slaughter (Santos et al., 2012; Gesualdi Junior et al., 2014).

Table 3
Fixed effects and their interactions of generalized mixed models fitted for ewes and lamb's behaviour soon after birth.

Ewe-lamb behaviour at birth	Dorper	Santa Ines	P-value		
			Breed	Litter size	Breed*litter size
Ewe's behaviour					
Labour duration (min)	75 ± 0.05	23 ± 0.04	0.0001	0.5260	0.8094
Latency to groom (s)	60 ± 0.09	60 ± 0.16	0.2890	0.0380	0.9507
Maternal grooming (%)	70 ± 0.20	90 ± 0.10	0.0745	0.0111	0.8555
Low-pitched bleats (n)	9 ± 0.20	14 ± 0.21	0.0008	0.1045	0.4788
Lamb's behaviour					
Latency to shake the head (s)	17 ± 0.09	4 ± 0.08	0.0030	0.6097	0.3584
Seeking the udder (%)	12 ± 0.14	17 ± 0.15	0.1073	0.9320	0.1174
Low-pitched bleats (n)	11 ± 0.10	6 ± 0.12	0.8500	0.7502	0.1120
Latency to stand (min)	13.5 ± 0.04	15 ± 0.05	0.3870	0.9724	0.8998
Latency to suckle (min)	49 ± 0.02	51 ± 0.03	0.9390	0.0702	0.3853

Means are depicted with ± SEM.

The lambs' growth and probability of survival until weaning rely on different factors, including birth weight, environmental conditions, milk/colostrum supply and the adequate expression of maternal care. Ewes display selective maternal care, exclusively nursing newborn lambs they bonded during the sensitivity period within the first 2 h after parturition (Poindron et al., 2007). After lambing, the ewes emit low-pitched bleats, licks and move around their lambs while they attempt to stand up (Nowak, 2006). Simultaneously, the lambs must stand up and move toward the udder to ingest colostrum as soon as possible (Nowak, 1996). During the first two weeks postpartum, the ewes allow their lambs to suckle as often as they want (Munro, 1956; Hinch, 1989), as mothers and lambs benefit from the maternal investment, a period during which the lambs largely depend on maternal care to survive (Weary et al., 2008). As lambs grow, ewes gradually start to prevent suckling bouts and are less vigilant, forcing lambs to gain more social and nutritional independence (Weary et al., 2008; Fonsêca et al., 2016).

The environment where the breeds were selected and adapted influences the behaviours of mothers and lambs at birth and during lactation, as evidenced, for instance, with Suffolk and Scottish Blackface ewes (Dwyer and Lawrence, 1997; Pickup and Dwyer, 2011). In the Brazilian semi-arid region, sheep production is based in hair breeds, and among these, Dorper sheep are used to produce heavier lambs (Santos et al., 2012). This augments the probability that Dorper mothers require assistance at delivery, negatively influencing newborn and maternal behaviours (Dwyer, 2003; Dwyer and Bünger, 2012; Regueiro et al., 2021, 2021). Additionally, as Dorper lambs grow faster than Santa Ines lambs (Santos et al., 2012), the former require more nutrients to grow, likely adapting their feeding strategy, leading to an advancement in the process of nutritional independence from their mothers. Considering all this information, we hypothesized that Santa Ines mothers establish and maintain a stronger attachment with their lambs at birth and throughout lactation. Since the characteristics of the bond between ewes and lambs are crucial factors explaining the lamb survival rate, this research might provide original insights into the components contributing to the high mortality rates of lambs from the widespread Brazilian breeds. Therefore, the aim of this study was to compare the behaviours of Santa Ines and Dorper ewes and their lambs at birth and throughout lactation.

Table 4
Fixed effects and their interactions of generalized mixed models fitted for ewes and lamb's behaviours throughout the lactation.

Ewe-lamb behaviour throughout the lactation	Dorper	Santa Ines	P-value				
			Breed	Weeks after parturition	Litter size	Breed*weeks after parturition	Breed*litter size*weeks after parturition
Ewe's behaviour							
Grazing (%)	60 ± 1.5	73 ± 2	0.3235	0.0141	0.6565	0.2939	0.8730
Lying (%)	37 ± 0.35	25 ± 0.25	0.0001	0.0031	0.0243	0.0756	0.7834
Head-up posture (n/4 hours)	1.5 ± 0.15	3.5 ± 0.2	0.0001	0.0001	0.0116	0.2333	0.0863
High pitched bleats (n/4 hours)	8 ± 0.2	12 ± 0.4	0.0196	0.0001	0.4517	0.1236	0.0623
Nose lamb (n/4 hours)	2.8 ± 0.1	4 ± 0.2	0.0001	0.0001	0.0516	0.8260	0.6213
Lamb's behaviour							
Grazing (%)	30 ± 1.5	50 ± 1.1	0.0001	0.0005	0.5350	0.4760	0.6240
Grazing close to mother (%)	27 ± 2.10	45 ± 2.8	0.0001	0.0003	0.1270	0.0820	0.8355
Lying (%)	45 ± 2	35 ± 1.8	0.0445	0.0480	0.1345	0.0775	0.8025
Lying close to mother (%)	42 ± 1.8	32 ± 1.5	0.0490	0.0535	0.8205	0.0250	0.9600
Within the creep feeding (%)	8.5 ± 0.5	7.5 ± 0.8	0.5665	0.0001	0.8580	0.8950	0.1945
Head-up posture (n/4 hours)	2.0 ± 0.2	2.5 ± 0.15	0.8982	0.0001	0.0320	0.2738	0.6665
Suckling attempt (n/4 hours)	9.5 ± 1.0	7 ± 1.20	0.1455	0.0001	0.6050	0.2090	0.8950
Suckling rate (n/4 hours)	6 ± 0.30	5.5 ± 0.10	0.7260	0.0001	0.5330	0.1160	0.7775
Suckling success (%)	60 ± 2.50	75 ± 2.60	0.0322	0.2319	0.2130	0.9200	0.9848

Means are depicted with ± SEM

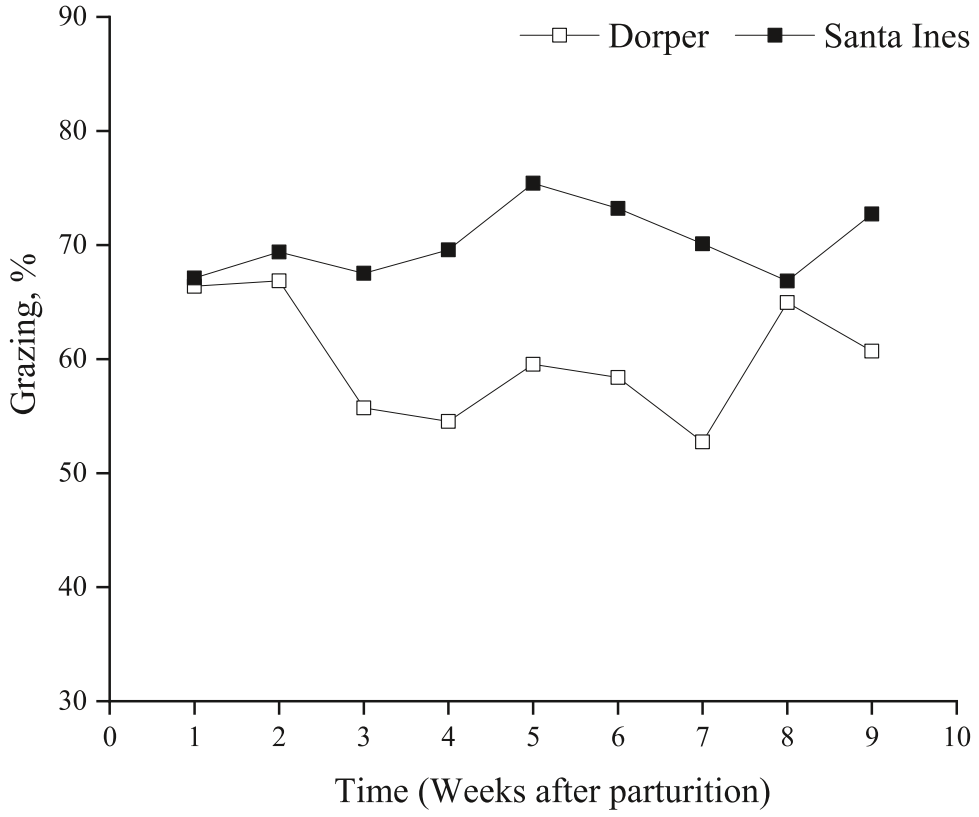

Fig. 1. Relative proportion of the time spent grazing of Santa Ines and Dorper ewes throughout the weeks after parturition. Mean difference for Dorper and Santa Ines ewes were not significant according to the pair-wise Tukey-Kramer test.

Furthermore, as single lambs have a stronger bond with their mother at birth and before weaning (Shubber et al., 1979; Nowak et al., 1989; O'Connor et al., 1992; Freitas-de-Melo et al., 2021), a complementary aim was to determine if being single or twin lambs impact differently on these breeds.

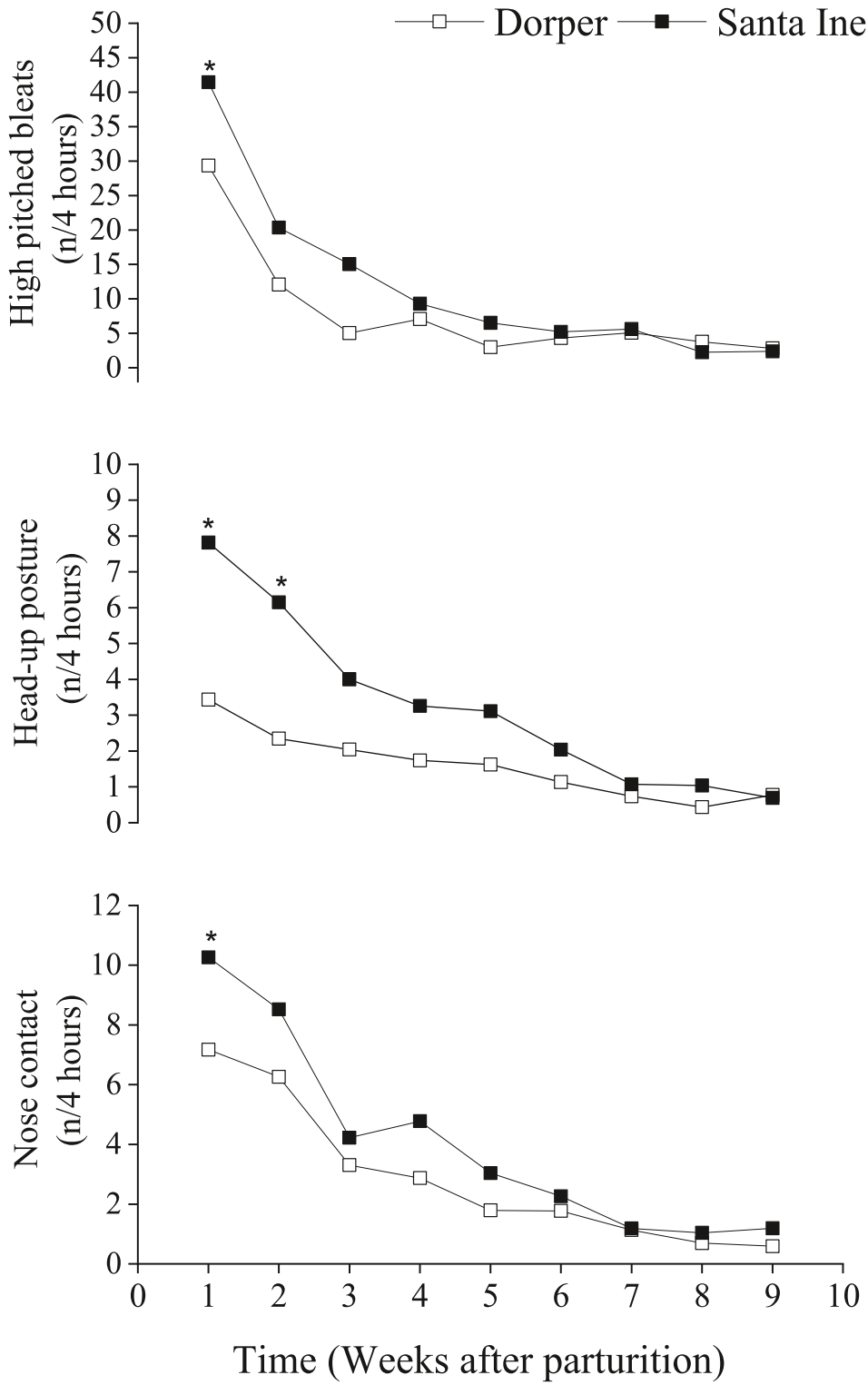

Fig. 2. Mean frequency of high-pitched bleats, head-up posture, and nose lamb expressed by Santa Ines and Dorper ewes throughout the weeks after parturition. The asterisk expresses significant ($P < 0.05$) breed differences by pair-wise Tukey-Kramer test.

2. Materials and methods

2.1. Location, animals and experimental design

The study was carried out at the Estação Experimental Benjamin Maranhão da Empresa de Pesquisa e Extensão Rural da Paraíba, Brazil (6° S, 35° W, 188 m altitude), between October and December of 2018 (spring in the southern hemisphere). Overall, climate classification according to Köppen-Geiger is BSh, semi-arid, with a mean annual precipitation rate below 500 mm year^{-1} (Alvares et al., 2014). All procedures involving animals were approved by the local committee of care and welfare of animals (CEUA-UFPB, 001174). Data were collected from 25 multiparous Santa Ines ewes and their single (n=18; females: 10; males: 8) and twin lambs (n=14; females: 8; males: 6) and 21

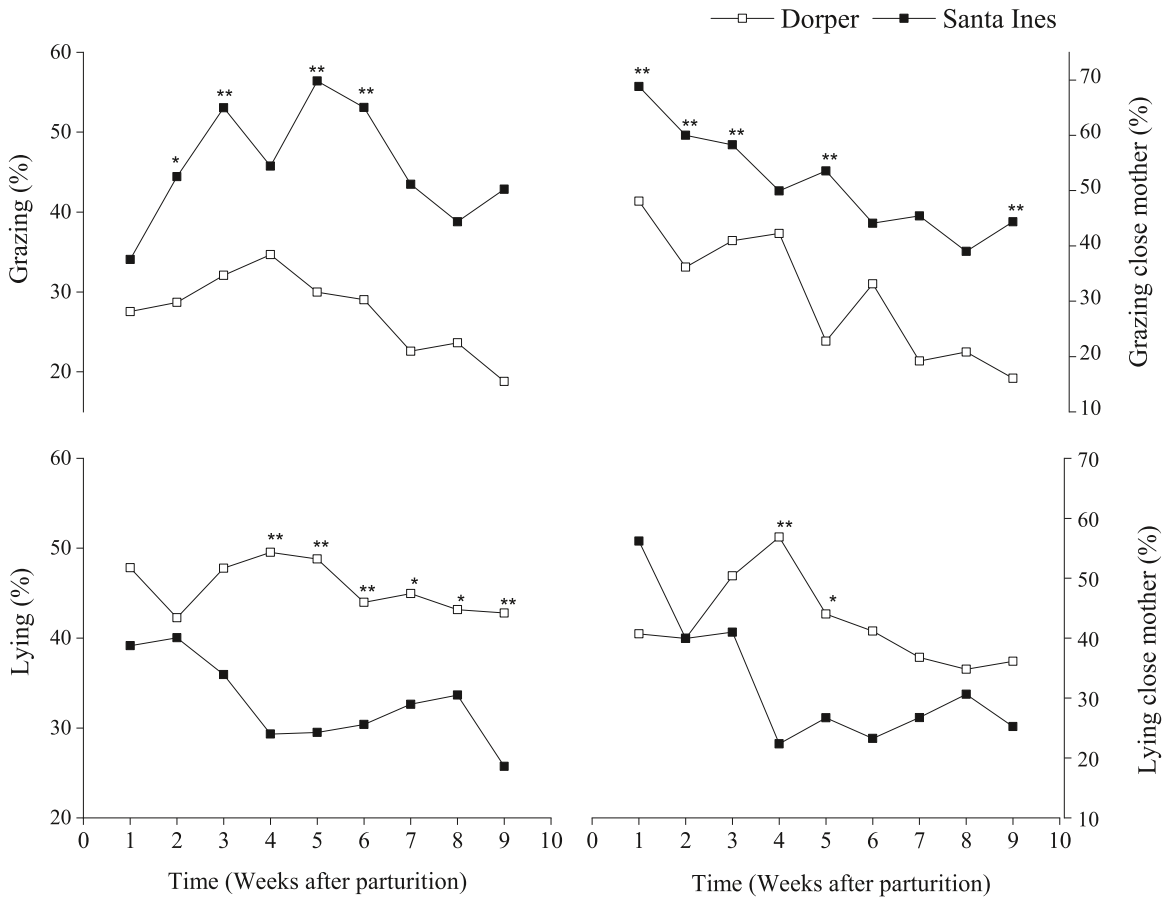

Fig. 3. Mean relative proportion of the time spent grazing, lying down, grazing and lying close to mother of Santa Ines and Dorper lambs over the weeks after parturition. Single and double asterisks express significant breed differences by pair-wise Tukey-Kramer test at $P < 0.05$ and at $P < 0.01$, respectively.

multiparous Dorper ewes and their single (n=16; females: 7; males: 9) and twin lambs (n=10; females: 4; males: 6).

2.2. General management during gestation, lambing season and lactation

During 42 days of the natural breeding season, ewes of each breed were joined with three fertile marking rams of the same breed painted on their chest [three black Santa Ines with a body mass of 77.5 kg (SD = 3.0), and three white Dorper with a body mass of 78.3 kg (SD = 2.0 kg)]. The day that ewes came into oestrus (Day 0) was identified by daily visual inspection of the coloured rumps of the ewes mated. Pregnancy and foetal number were confirmed by transrectal ultrasonography approximately 35 days after oestrus. During the breeding season and throughout the gestation, ewes remained in an outdoor paddock (0.8 ha), grazing *Panicum maximum cv. Aruana,* from 06:00 to 17:00 h. The paddock had availability of natural shade projected by *Prosopis juliflora* and *Ziziphus joazeiro*. During the rest of the day (from 17:01 to 5:59 h), all animals were housed in roofed pens with earthen floor, where they were fed with corn, cotton cake, wheat bran, limestone and mineral mixture, adjusted for a mean daily intake of 300 g per animal, according to the NRC (2007). Mineral supplementation (Ovinofós, DSM Tortuga, Brazil) and water were freely available. Approximately five days before the expected parturition date (average: 145 days of gestation), all ewes were moved to a lambing pen (16 m × 7 m), where the ewe-lamb behaviours were continuously recorded during the whole 24 h period by trained observers until all ewes lambed (see more details below). During this period, in addition to concentrate supplementation, also adjusted for a mean daily intake of 300 g per animal (NRC, 2007), ewes were collectively fed in wood feeders (0.30 m × 0.20 m × 12 m, width × depth × length) with 0.5 kg of DM animal^{-1} day^{-1} of roughage, of which 20 % was Tifton hay and 80 % was spineless cactus (*Nopalea cochenillifera*). The lambing pen was also with earthen floor, surrounded by wood fences, and having shelter from two roof structures of fibre cement (2 m × 8 m × 2 m, 2 m × 7 m × 4 m, height × length × width respectively), with a slope close to 15 %. Ewes and lambs remained in the lambing pen until three days after birth. Thereafter, ewes and lambs were moved again to the outdoor paddock, where ewes had been during gestation. In the paddock, lambs had access to a total mixed ration (23.4 % of crude protein and 3.0 Mcal of metabolizable energy) in a creep-feeding (3 m × 2 m, length × width) based on Tifton hay, corn meal, soybean meal, soy oil, mineral salt, and calcitic limestone. The feed was provided in a collective wood feeder (0.2 m × 2 m × 0.3 m; depth × length × width), placed within the creep-feeding.

2.3. Mother-offspring behaviours at birth

The lambing season lasted 36 days (from October 6 to November 11 of 2018). Ewes were numbered with nontoxic paint on the right and left sides to facilitate identification by the observers. Continuous focal sampling of the behaviours of ewes and their lambs were performed by ten trained observers from a distance enough to avoid disturbing the normal behaviour of animals. Before starting the behavioural observations, the observers were subjected to an inter-observer agreement trial using 12 hours of video footage from deliveries recorded during the study of Fonsêca et al. (2016), until they achieved ~95 % similarity of behavioural measures among observers. The lambing pen was kept overnight without any source of artificial light. Latency for the ewe to groom the lamb, the number of low-pitched bleats and the duration of grooming were recorded. Latency for the lamb to shake its' head, stand up and suckle, the number of low-pitched bleats and the duration of

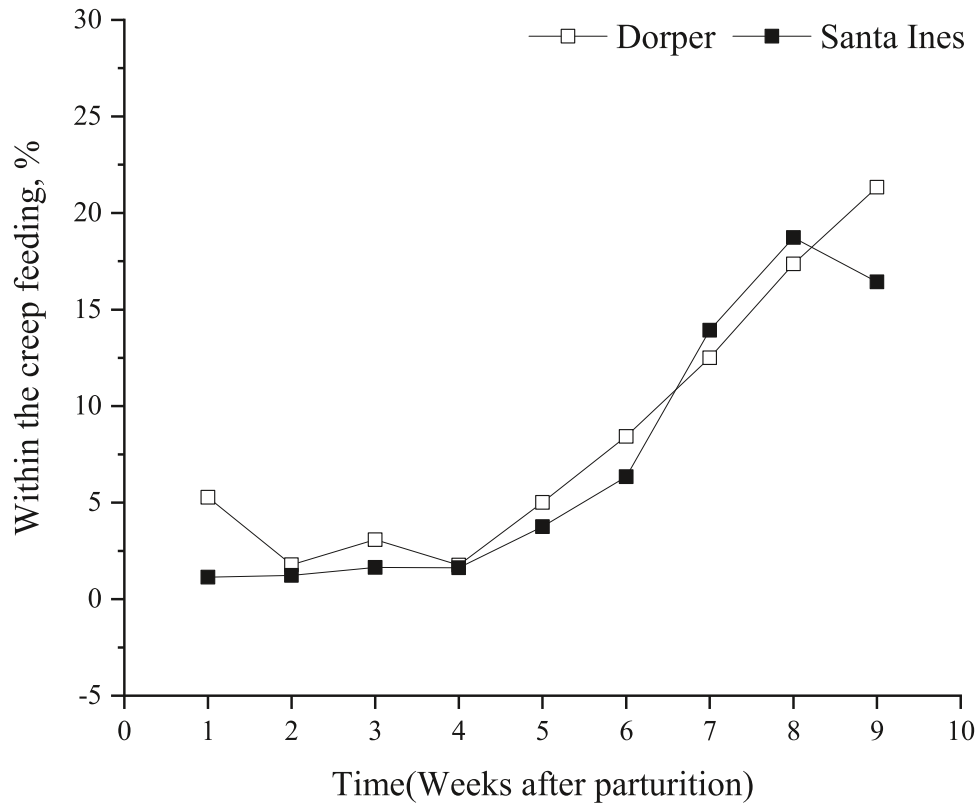

Fig. 4. Mean relative proportion of time spent within the creep-feeding by Santa Ines and Dorper lambs throughout the weeks after parturition. Mean difference between Dorper and Santa Ines ewes were not significant ($P > 0.05$) according to the pair-wise Tukey-Kramer test.

seeking the udder were registered. The behaviours of ewes and lambs recorded at birth are described in Table 1. The duration of birth labour was recorded considering the lapse from the appearance of fluids until the moment that the birth of the last lamb ended (Dywer et al., 2003). It was considered that the birth was dystotic when the foetal front or rear legs appeared at the vulva with no progress after 1:30 h. This occurred on six occasions, all in Dorper lambs, so human assistance was provided for ending the birth. Mother-offspring behaviours were recorded from the end of the second stage of parturition until 2 h after birth or at the moment of the first successful suckling by the lambs. Lambs that had not suckled after 2 h after birth were assisted to suckle, and the latency to suckle for the first time was considered as 120 min. The human assistance to suckle was required in one newborn delivered from a Santa Ines ewe and three newborns delivered from Dorper ewes. At 4 h after birth, the lambs were weighed, tagged, and their sex registered.

2.4. Ewe-lamb behaviour throughout the lactation

The behaviours of ewes and lambs were continuously recorded on days 7, 14, 21, 28, 35, 42, 49, 56 and 63 after lambing, from 06:00 h to 10:00 h, using focal individual sampling (Lehner, 1996). As the lambing season lasted 36 days, the number of ewes and lambs recorded each day depended on the lambing date. The biggest group evaluated in a single day during this period had seven ewes and eight lambs, while the smallest had five ewes and their nine lambs. Ewes and lambs were individually numbered with nontoxic black paint on their right and left sides. Ten trained observers recorded the duration and the frequency (number of occurrences per unit of time) of ewes' and lambs' behaviours. Each observer recorded the behaviour of one ewe and their lamb (s). Observers were also trained using 12 hours of video footage from evaluations performed in the study of Fonsêca et al. (2016). They were kept, as much as possible, at a distance of 10 m apart from ewes and lambs, recording them with the aid of binoculars and stopwatches. The observations were performed without disturbing ewes, as they were accustomed to human handling procedures. Before starting the behavioural observations, the observers were subjected to an inter-observer agreement trial in which they also achieved ~95 % similarity of behavioural measures among observers. The times that ewes spent grazing and lying down, the number of high-pitched bleats, head-up posture, and nose contact were recorded. The time that lambs spent grazing, lying down, within the creep-feeding, close to their mother, and the number of high-pitched bleats, head-up posture, suckling attempts and successful suckling were recorded (Fonsêca et al., 2016). Moreover, the percentage of successful suckling was determined as follows: (suckling attempts/suckling rates)*100. The description of ewes' and lambs' behaviours is depicted in Table 2.

2.5. Statistical analyses

All data were analyzed using SAS System (SAS on demand for academics, Inst. Inc., Cary, NC). Data were first tested for normality with the Shapiro-Wilk test, which attested asymmetric distribution for all behavioural variables. The variables described in Tables 1 and 2 were compared with generalized mixed models (Proc Glimmix) fitting lognormal distribution for variables measured as time duration, and negative binomial for variables measured as frequencies. Except in labour duration, and the latencies to groom, head shaking, to stand up and to suckle, all the behaviours that were measured as time duration had their relative proportion of time calculated. For the data recorded at birth, the model included the breed (Santa Ines and Dorper) and the litter size (singletons and twin lambs) as the main effects, as well as the interaction between breed and litter size. The relative frequency of dystocia for Dorper and Santa Ines was compared by the Fisher's exact test. For ewe-lamb variables recorded throughout the lactation, the model included the breed (Santa Ines and Dorper), the litter size (singletons and twin lambs) and the time (weeks after birth) as the main

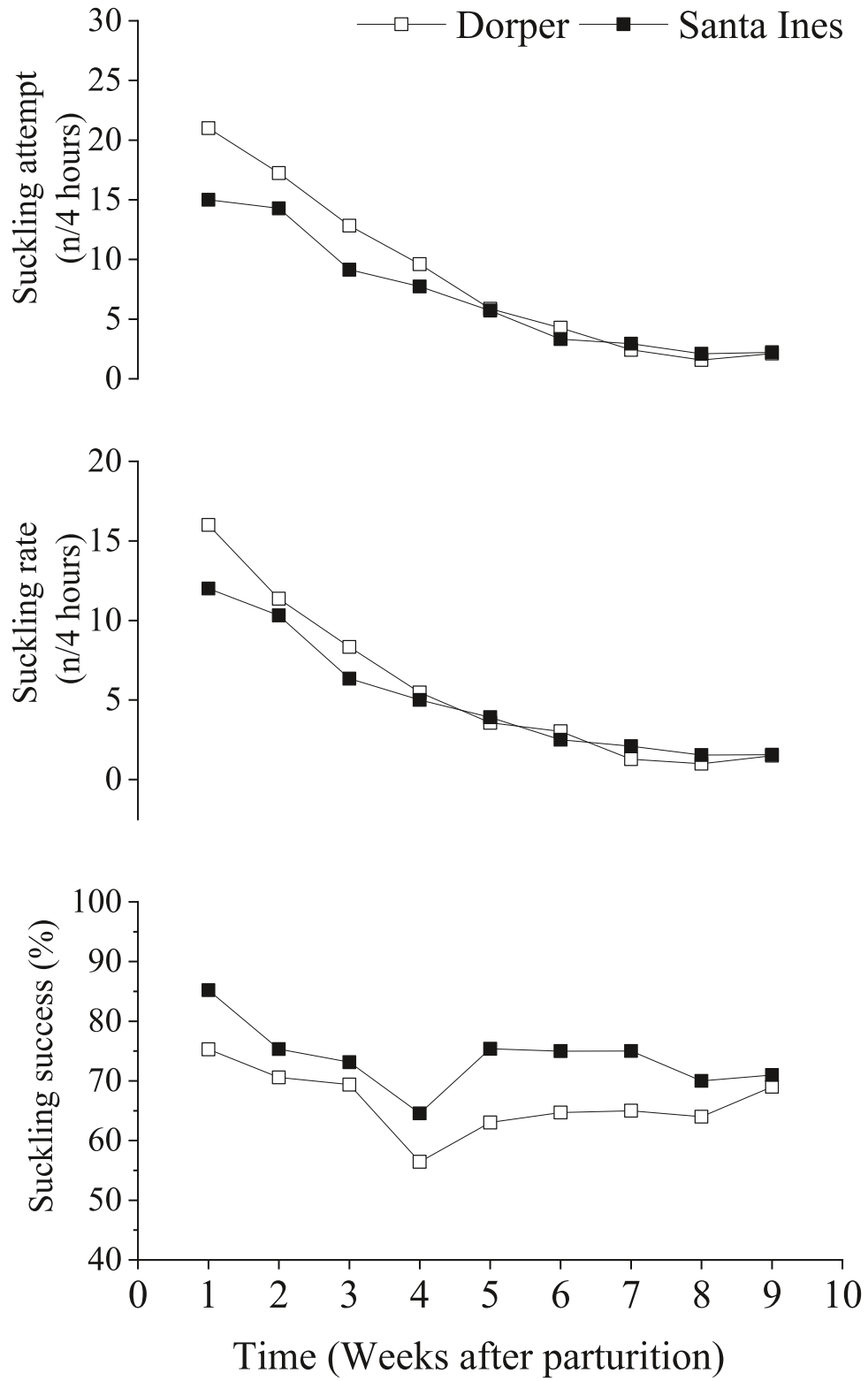

Fig. 5. Mean frequency for suckling attempt, suckling rate, and mean proportion of suckling success of Santa Ines and Dorper lambs. Mean difference between Dorper and Santa Ines ewes were not significant ($P > 0.05$) according to the pair-wise Tukey-Kramer test.

effects, in addition to their interactions. Pair-wise comparisons were made using the Tukey-Kramer test. Differences were considered as significant when $P \leq 0.05$, and as tendencies when $0.05 < P \leq 0.1$. Data are presented as back-transformed means.

3. Results

3.1. Birth weight and mortality rate

The breed did not affect the lambs' birth weight: Dorper (n = 26; 3.80 kg [SD = 1.5]); Santa Ines lambs (n = 32; 3.67 kg [SD = 1.4]).

However, single-born lambs (n = 34; 4.12 kg [SD = 1.2]) were heavier than twins lambs (n = 24; 3.35 kg [SD = 1.1]) at birth. During the study, two of 26 Dorper lambs died (mortality rate of 7.7 %), while two of 32 Santa Ines lambs died (mortality rate of 6.3 %).

3.2. Ewe-lamb behaviour at birth

There were no significant interactions between breed and litter size for all ewes and lambs' variables (Table 3). Labour in Santa Ines ewes was shorter than in Dorper ewes ($df = 56$; $F = 20.63$; $P = 0.0001$) (Table 3). Likewise, the incidence of dystocia was greater in Dorper than Santa Ines ewes ([18 % vs. 6 %]; $P = 0.018$). Although the breed did not affect the latency to groom, Santa Ines ewes displayed a greater frequency of low-pitched bleats ($df = 56$; $F = 13.11$; $P = 0.0006$) and tended to spend more time grooming their lambs ($df = 50$; $F = 3.32$; $P = 0.07$; Table 3). The Santa Ines lambs shook their heads earlier than Dorper lambs ($df = 56$; $F = 10.23$; $P = 0.003$), but no differences were detected in latencies to stand up and to suckle. Ewes of single deliveries started to groom their lambs earlier than those that lambed twins ([22 s vs. 90 s] $df = 50$; $F = 4.60$; $P = 0.04$), and those that delivered twins spent more time grooming the first lamb delivered ([90 % vs. 70 %] $df = 50$; $F = 6.97$; $P = 0.01$), independently of the breed (Table 3). Moreover, single-born lambs also tended to suckle earlier than twins ($df = 56$; $F = 3.41$; $P = 0.07$) (Table 3).

3.3. Ewe's behaviour throughout the lactation period

The time that ewes spent grazing did not differ between Santa Ines and Dorper ewes (Table 4; Fig. 1). Over the first two weeks after parturition, the Santa Ines ewes displayed higher frequencies of head-up postures ($df = 412$; $F = 14.90$; $P = 0.0001$), high-pitched bleats ($df = 412$; $F = 6.49$; $P = 0.02$) and nose contacts ($df = 412$; $F = 23.40$; $P = 0.0001$) than Dorper ewes (Table 4; Fig. 2). Overall, these behaviours decreased ($P < 0.05$) in frequency along the lactation phase independently of the breed. In terms of litter size, the head-up posture and nose contact were more frequent in mothers that delivered single than twins lambs independently of the breed ($df = 412$; $F = 8.50$; $P < 0.05$) (Table 4).

3.4. Lamb's behaviour throughout the lactation period

The Santa Ines lambs devoted greater time grazing than Dorper lambs over the weeks of observation ($df = 1758$; $F = 29.56$; $P < 0.0001$), while the opposite occurred for the time spent lying down ($df = 1758$; $F = 8.20$; $P = 0.05$) (Table 4; Fig. 3). The Santa Ines lambs were closer to their mother when grazing ($df = 1908$; $F = 13.42$; $P = 0.0001$), while Dorper lambs were closer to their mother when lying down ($df = 1156$; $F = 3.92$; $P = 0.049$) (Table 4; Fig. 3). Independently of the breed, the time spent grazing and lying down close to the mother decreased with age, while the time spent within the creep-feeding increased ($P < 0.05$; Table 4; Figs. 3 and 4). Similarly, the number of suckling attempts and effective sucklings decreased with lamb's age ($P < 0.05$; Table 4; Fig. 5). With no difference according to the breed, a higher proportion of successful suckling (80 %, on average) occurred in the first week since birth, decreasing progressively ($P < 0.05$; Table 4; Fig. 5) until the week four. After that, it remained relatively stable until the end of the study for both breeds. Lastly, the effect of litter size and the interaction between breed and time after birth were not significant for any lamb's variables analyzed (Table 4).

4. Discussion

To the best of our knowledge, this is the first long-lasting study that compared temporal changes in the mother-offspring relationship in two hair-coat sheep breeds. In general, the results confirmed the initial hypothesis: Santa Ines, a sheep breed naturalized to the Brazilian equatorial semi-arid conditions, displayed a stronger ewe-lamb bond than Dorper, an introduced exotic breed. Three main findings support this statement. First, Santa Ines ewes displayed a higher frequency of low-pitched bleats at birth - a main indicator of maternal motivation - than Dorper ewes and devoted greater time grooming their lambs, who were quicker to shake their head after birth. Second, the expression of vigilant and affiliative behaviours, such as high-pitched bleats, head-up posture and nose contact, were more frequent in Santa Ines than in Dorper ewes during the first two weeks postpartum. Lastly, from the first week after lambing until around 2 months of age, Santa Ines lambs remained closer to their mothers when grazing than Dorper lambs. These results extend previous investigations performed with sheep from temperate latitudes, which show that breeds less selected for sheep production display a stronger mother-offspring bond than those more intensively selected for meat production (Dwyer and Lawrence, 1997; Pickup and Dwyer, 2011). Moreover, with these results, it would be interesting to study the role of the lambs in the establishment and evolution of the bond to disentangle the role of the mother and the lamb (Stevens et al., 1984). It should be considered, also, that while Dorper sheep was introduced to Brazil expecting to enhance meat production, Santa Ines breed was developed in environments similar to that used in the study, therefore, being probably better adapted to the local conditions.

The maternal behaviour of Dorper ewes at birth could have been worsen by their longer labour and greater incidence of dystocia than Santa Ines ewes. Extended duration of parturition and the occurrence of dystocia impairs maternal and newborn behaviours at birth, reducing the ewes' motivation to lick the lamb and the lambs' vigour due to asphyxia (Dutra and Banchero, 2011; Regueiro et al., 2021). Accordingly, Dorper ewes licked their lamb for a shorter time at birth, and their lambs took longer to shake heads after birth than those delivered by Santa Ines mothers. It is interesting to note that the longer duration of labour in Dorper ewes was indeed not caused by lambs' birth weight, as previously reported (Regueiro et al., 2021). Similarly, the greater incidence of dystocia in Dorper than Santa Ines ewes was unrelated to lambs' body weight. It is worth to speculate about the existence of differences between breeds in lambs' thorax circumference, which is positively correlated with dystocia incidence (Jacobson et al., 2020), usually greater in meat-specialized breeds, such as Dorper (Figueiredo et al., 2019). Dystocia is an important cause of lamb mortality and increases the likelihood of postpartum ewe infection, leading to economic losses for farmers and raising animal welfare concerns (Jacobson et al., 2020). Therefore, these results highlight the necessity of more studies aiming to understand the determinants of the differences in the duration of labour and the incidence of dystocia in Dorper and Santa Ines ewes.

Breed differences concerning the level of attachment between ewes and lambs in the immediate postpartum persisted over the lactation. During the first two weeks postpartum, Santa Ines ewes displayed a greater frequency of high-pitched bleats and head-up posture than Dorper ewes. The Santa Ines lambs spent greater time close to their mother when grazing than Dorper lambs. During the first week of life, even though lambs are not fully developed to digest and absorb nutrients derived from solid food, a close spatial ewe-lamb relationship is essential for the lamb to establish food preferences, avoid ingesting toxic compounds, and acquire grazing skills (Mirza and Provenza, 1992). Moreover, in this period the mother is the main promoter of the lamb socialization and learning of the behavioral pattern of the species. The expression of vigilant behaviours by mothers and the maintenance of a close spatial ewe-lamb relationship in the first week postpartum plays therefore high adaptive value to the lambs' fitness, especially in those raised in extensive production systems, where human assistance is marginal and environmental challenges are greater than in intensive conditions. In this study, although animals had access to a homogeneous pasture, breed differences detected may also translate the historical evolution process of Santa Ines sheep under extensive production systems, where sheep typically graze on heterogeneous pastures such as the

Caatinga biome, a tropical savanna (dos Santos et al., 2023). Previous findings with woolly sheep also attested that the Scottish Blackface, a less artificially selected breed for wool production, were more vigilant and closer to their lambs than Suffolk sheep, a more intensively-selected breed (Dwyer and Lawrence, 1998; Pickup and Dwyer, 2011). Thus, it should be interesting to determine in bigger flocks if this difference is related to a greater protection from predators in the Santa Ines breed.

The suckling attempts and the suckling events decreased during the studied period in both breeds. From the first week postpartum, the proportion of successful suckling averaged between 70 – 80 %, indicating that ewes started to control rates of milk transfer very early by preventing some suckling bouts by lambs. While this result challenges the idea that during the first two weeks postpartum, ewes allow their lambs to suckle *ad libitum* and do not restrict access to the udder (Ewbank, 1967), it strongly confirms that the weaning process in sheep occurs smoothly, where gradually lambs' milk dependence decreases (Lee et al., 1991). This process induces lambs to seek other sources of nutrients, which can influence an earlier maturation of the digestive tract, increasing their capacity to digest solid foods. Our results evidenced that the time spent grazing increased only in Santa Ines lambs, but the time spent within the creep-feeding increased in general, despite the breed. Similarly, Santa Ines × Dorper crossbred lambs raised under pasture-based system, and with free access to creep-fed progressively increase the nutritional independence from their mothers by the reduction of time suckling and the increase in the consumption of concentrate within the creep-fed (Melo et al., 2019). However, although the greater time in the creep-feeding area can indicate nutritional maturation, it can also be related to the strength of the relationships between the lambs, and therefore, a stronger gregariousness pattern among the lambs as they gain independence from their mothers. The different strategies on how lambs gain weight should be considered beyond the body or the carcass weight itself to develop specific strategies to maximize the potential of each breed, an issue that should be addressed in future studies.

In this study, both for Santa Ines and Dorper sheep, the most apparent phase of disagreement between mothers and lambs occurred within week four. From this point, the proportion of successful suckling increased and remained relatively stable over the rest of the weeks. Overall, the dynamics of mother-offspring relationship throughout the lactation period observed in this study were similar to those reported with woolly sheep breeds and free-ranging sheep from subtropical and temperate climates (Merino sheep, Arnold et al., 1979; Mountain Bighorn Sheep, *Ovis canadensis*, Berger, 1979; Scottish Blackface and Suffolk, Pickup and Dwyer, 2011), in addition to hair sheep breeds from tropical areas (Saint-Croix sheep, Maldonado et al., 2015; Morada Nova sheep, Fonsêca et al., 2016). These authors also attested that the strength of maternal care is higher over the first weeks after birth, and progressively decreases with the increase in the social and nutritional independence of lambs from their mother.

Despite the breed, maternal behaviour was stronger toward single than twin lambs (e.g., time grooming the lamb and the frequency of low-pitched bleats), and single lambs tended to ingest colostrum earlier than twins. Interestingly, the lack of breed differential effects on single or twin lambing ewes allows to consider the increase of the litter size as a productive strategy per se, with similar potential effects despite the breed used. However, it is important to point out that in this study ewes and their lambs remained in close contact during the period immediately after birth, promoting the bonding, an issue that might differ from what is observed when ewes deliver and rear their lambs in extensive grazing paddocks. There are several nutritional or hormonal strategies to increase the ovulation rate, but it should be considered that in extensive systems, the greater mortality rate of twins can be partially explained by the differences in maternal bond reported in this study. In this sense, in previous studies it has also been confirmed that ewes and their twin lambs develop a weaker bond than ewes of single lambs in the early postpartum (O'Connor et al., 1992; Nowak et al., 1989), and this pattern persists throughout the lactation (Hinch, 1989). The ewes that delivered twin lambs have to split attention and nursing between two lambs rather than a single one, partially explaining the weaker mother-young bond in twins. In this study, although ewes that delivered single lambs were more vigilant during the lactation than those that delivered twins, there was no strong evidence that ewes and their twin lambs had a weaker bond during this phase. In this sense, our results show that temporal changes in ewe-lamb bond in terms of spatial relationship and suckling activity were more influenced by breed differences instead of litter size. However, in previous studies it was reported that the weaker bond of twins persists until weaning, as their responses differ after abrupt separation (Freitas-de-Melo et al., 2021; Freitas-de-Melo et al., 2022).

In conclusion, Santa Ines sheep, the local evolved breed, developed a stronger mother-offspring bond than Dorper from birth until at least two months after lambing. This highlights the importance of selection adapted to local environmental conditions, as this impacts traits that are not frequently considered but can have a great impact on productive results such as maternal behavior. However, the dynamics of mother-offspring disagreement throughout the lactation did not differ between Santa Ines and Dorper sheep. Ultimately, the ewe-lamb bond was weaker in twins than in single lambs, without differences in the degree of its strength according to the breed.

Ethics approval

All procedures involving animals were reviewed by the local committee of care and welfare of animals (CEUA-UFPB, 001174).

CRediT authorship contribution statement

Vinicius França Carvalho Fonsêca: Writing – review & editing, Writing – original draft, Visualization, Validation, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Edilson Paes Saraiva:** Resources, Project administration, Methodology, Funding acquisition, Conceptualization, Supervision. **Larissa Kellen Da Cunha Moraes:** Visualization, Methodology, Investigation. **Edgard Cavacanti Pimenta Filho:** Resources. **Wandrick Hauss de Sousa:** Resources. **Geni Caetano Xavier Neta:** Methodology, Investigation. **José Danrley Cavalcante Dos Santos:** Visualization, Methodology, Investigation. **Aline Freitas-de-Melo:** Writing – review & editing, Visualization, Validation, Supervision, Methodology, Data curation, Conceptualization. **Rodolfo Ungerfeld:** Writing – review & editing, Visualization, Validation, Supervision, Methodology, Data curation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

We ackonowledge the National Council of Scientific and Technological Development (CNPQ) for the grant received (Proc. Number: 420903/2016-0).

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