



In the Absence of Sexual Competition, Rams with Low Social Ranks Show more Appetitive Sexual Behaviors than Males with High Ranks

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ABSTRACT

Background: Usually, social order is defined by the animals' size, live weight, or age. This study evaluated the effect of social rank and morphometric characteristics upon appetitive and consummatory sexual behaviors in Dorper rams in absence of sexual competition.

Methods: The research took place in northern Mexico (26°N), 36 rams allocated into two groups were used: Low social rank (LR; n = 17) and high social rank (HR; n = 19). Behavior tests were performed for all the males, for which each one was placed in contact with a female in estrus.

Result: There were no differences between ranks and morphometric characteristics ($P < 0.05$). A correlation of 29% was found for the success index concerning odor in HR. Likewise, a relationship existed between the presence of horns (Pho) and all the other variables, but not between PHO vs success index ($\rho = 0.07$). Appetitive sexual behavior was the highest ($P < 0.05$) for LR, while HR had more consummatory sexual behaviors ($P < 0.05$). We conclude that sexual hierarchy exists even when the males have the same morphometric characteristics, nonetheless, males with low social ranks show more appetitive sexual behaviors while those with high ranks show a higher number of consummatory sexual behaviors when managed in intensive farming conditions.

Key words: Appetitive, Consummatory, Ram, Sexual behavior, Social rank.

INTRODUCTION

Domestic sheep are very important for humans, due to their nutritional and productive value (Flota-Bañuelos *et al.*, 2019). Sheep have social structures that are of significance from the productive and reproductive points of view (Martin *et al.*, 2013), their social organization is in a hierarchical order that consists of dominant and subordinate animals (Díaz *et al.*, 2021), which can determine the relations between behavioral and secondary sexual traits (Maksimovico *et al.*, 2012). The hierarchical differentiation begins at an early age, affecting and determining the behavioral and morphological development in their adult lives (Ungerfeld and Gonzalez-Pensado, 2008), with a higher body growth rate, more behavioral and morphological developments are observed in dominant males (Pelletier and Festa-Bianchet, 2006). This occurs mainly when the animals are managed in extensive conditions. However, this has not been thoroughly researched in intensive conditions, where there is enough feed for them to develop their body and sexual characteristics. On the other hand, very little is known about the behavior of rams of low hierarchy with females in estrus without the sexual competition of dominant males, because most of the research has been implemented with competition between them (Orihuela Trujillo, 2014). Historically, most of the hierarchy research has been performed in wild-life extensive systems, where subordinate males generally have less access to feed, which affects their morpho-

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logical development and, in consequence, their sexual development (Fournier and Festa-Bianchet, 1995). Thus, the reproductive capacity of low hierarchy males could be underestimated, in an environment free of competition and intensive management systems. Also, a new characterization of sexual behaviors of rams when exposed to females in estrus has recently been carried out, which has been improved and the number of sexual behaviors has been expanded and divided into appetitive and consummatory sexual behaviors, besides stress be-

aviors (Tejada *et al.*, 2016; Calderín-Leyva *et al.*, 2018). Therefore, it would be interesting to determine if the social rank affects these types of behaviors in animals without sexual competition and bred in intensive conditions because it is probable that low social ranking males are being underestimated when they could be a good alternative for producers. After all, they have an adequate body and sexual development and a more docile behavior than high hierarchy males. Thus, this study aimed to evaluate the effect of the morphological characteristics upon social rank and sexual behavior of Dorper rams without sexual competition, under intensive management systems.

MATERIALS AND METHODS

All methods and management of experimental units used in this study were in strict accordance with the guidelines for the ethical use, care and welfare of animals in research at the international (FASS, 2010) and national (NAM, 2002) levels.

Study site and environmental conditions

The study was carried from March 21 to May 7, 2017, in Matamoros, Coahuila, Mexico, in the commercial farm “El milagro” (25°LN 103° LO; 1,120 mams). The area has dry climate, with temperatures between 9.20 and 28.3°C (INIFAP LA LAGUNA, 2019).

Experimental animals and their management

Thirty-six males and 10 multiparous Dorper females on average 3 years old and fed twice daily (10:00 h and 18:00) with leftover cattle feed (17% CP and 1.5% ME), were selected. The animals had free access to water and mineral salt, received vitamins and were dewormed 3 weeks before the study also, their health status was reviewed by a veterinarian.

Social ranking evaluation (Success index)

The social rank of each male was determined by a behavioral test adapted from Alvarez *et al.* (2003) and Barroso *et al.* (2000). Each male was confronted with the rest of the males. The won and lost events of each male were determined by trained technicians. The success index (SI) was the result of dividing the number of won events by the number of lost events. The social rank of rams was evaluated using ten estrogenized ewes. Rams were exposed to ewes in individual 2.5 × 2.5 m pens. In each behavioral evaluation, two males were formed until each ram competed against the remaining ten rams. A ram-to-ewe interaction was considered when a ram displayed dominant behaviors towards the other ram and declined the interaction (subordinate male). To define the success rate, the behavioral characteristics linked to success from the ram-to-ram competitions were considered; in sheep, the opportunity for male reproduction is primarily determined through paired-male rounds. Each ram fought all the other rams. Also, mounts with ejaculation and rejection were evaluated. The observations of the behavioral study (*i.e.*, agonistic interactions) were made by a single person and the

information was recorded in a digital format. Then, the SI for each ram was quantified determining two social ranks: low (LR; SI = 0-0.49) and high (HR; SI = 0.50-1.0).

Head morphometric characteristics and body temperature of the animals

The morphometric characteristics of the rams were based on the following aspects: body weight, head length (HeL; cm), head width (HeW; cm), horn length 1 (HoL1; cm), horn length 2 (HoL2; cm), horn width (HoW; cm), total horn length (THoL; cm), presence of horns (PHo) and absence of horns (AHo). These measurements were recorded with the use of a flexometer with a minimum of 0 and a maximum of 150 cm.

Sexual behavior evaluation

After the social ranking evaluation, behavior tests were performed on the 36 males, which were divided into two groups based on their social ranking: low rank (LR; n = 17) and high rank (HR; n = 19). The sexual behavior test consisted in exposing each male to a female in estrus in individual pens of 2.5 × 2.5 m, during 15 min, the frequency of each sexual behavior was recorded; appetitive (ASB; anogenital sniffing, approximations) and consummatory (CSB; mount attempt, full mount, mount with ejaculation). Also, the temperament behavior (TEMP) was evaluated by considering the frequency of a) flight attempt (number of times de male tried to escape from the pen), b) isolation (number of times that the male stayed still one meter or more away from the female, without showing any behavior) and c) aggression (number of times that the ram butted the ewe).

Statistical analysis

Data was previously analyzed by a “Shapiro test” to determine normality. Similarity analysis was performed with “metaMDS” for the morphometric characteristics according to the social rank with 1000 permutations and a Stress index of 0.0120. Likewise, a “permanova” test was performed to determine differences between ranges and morphometric characteristics. Subsequently, a Spearman correlation test was performed on each social rank using “chart”. Correlation to determine the relationship between the success index with morphometric characteristics. Behavioral values were analyzed by the general linear model “glm”. The statistical model to analyze the response variables include the social rank effect, results are shown as mean ± SE and are considered statistically significant at P<0.05. All the procedures were executed with the R program version 4.0.5 (2021-03-31).

RESULTS AND DISCUSSION

There was no significant difference from rams of LR vs HR regarding weight (83.4±3.6 vs 81.1±2.0; P>0.05, respectively) and BCS (3.2±0.1 vs 3.2±0.1; P>0.05, respectively). In Table 1, won events, lost events, success index, mount with ejaculation and rejections are shown. There were no differences between both ranks for libido variables (P>0.005). Nonetheless, won events were lower than lost events for LR with an average of 36% and 64% (P<0.05), respectively, while the

HR was inversely proportional, that is, the average for won events was 62% and 38% for lost events ($P < 0.05$). the HR showed a higher success index than the LR, $P < 0.05$.

Morphometric characteristics of rams

In Table 2, morphometric characteristics of Dorper rams with different social ranks are shown. On the other hand, there was no difference between the rest of the morphometric characteristics (HeL, HeW, HoL 1, HoL 2, HoW, THoL, $P > 0.05$).

In Table 3, the appetitive and consummatory sexual behaviors and the stress temperament of rams from both social ranks are shown. Males from the LR showed more ASB when compared to males of HR ($P < 0.05$). However, males from HR showed more CSB than LR ($P < 0.05$). Nonetheless, TEMP was similar between males from both ranks (0.3 ± 0.2 ; $P > 0.05$).

Fig 1 shows the correlation index between LR and HeL, HeW, HoL1, HoL2, HoW, THoL, odor, Pho and AHo. There was no correlation found between the index and morphometric characteristics, however, there was a correlation found within morphometric characteristics, that is to say, there was a relation between HeL and HeW, HoW, odor and Pho.

Fig 2 shows the correlation between the index from HR and HeL, HeW, HoL1, HoL2, HoW, THoL, odor, Pho and AHo. Index and odor were 29% related. Likewise, a relation between Pho and the variables odor, THoL, HoW, HeW and HeL. No relation was found between Pho vs index ($\rho = 0.07$).

In the present study, we found a difference between the social ranges of Dorper rams even though the morphometric characteristics were similar. Indeed, the males of the present study in both groups had similar weight, body condition and morphological measures ($P > 0.05$). The level of hierarchy was not affected by their body development. However, in high-ranking animals, there was a correlation between success index and odor. Therefore, high-ranking males had higher odor levels than low-ranking males, this could indicate that these animals had a higher level of testosterone. Although in our study testosterone levels were not measured, we did measure odor, which is directly related to the levels of this hormone (Calderón-Leyva *et al.*, 2018). Even though testosterone is normally related to sexual behavior (Pelletier and Festa-Bianchet, 2006), it is possible that low hierarchy males had enough levels of testosterone to express sexual behavior. It has been stated that testosterone increases with age and it is not related to the social ranking of growing lambs (Ungerfeld and González-Pensado, 2008). There may also be a pheromone factor. Female odor stimulates luteinizing hormone (LH) and testosterone secretion in males (Cohen-Tannoudji *et al.*, 1994). Ungerfeld *et al.* (2006) say that the pheromone stimulus of estrous ewes determines an increase in ram's sexual performance.

We observed that males of different hierarchies expressed different sexual behaviors, in this sense, the high ASB of the males in the present study could be that low hierarchy males had a level of testosterone that was sufficient to express good sexual behavior, however testosterone was

Table 1: Social ranking determination of Dorper rams exposed to estrus females.

Rams (n)	LR (17)	HR (19)
Libido		
Mounts with ejaculation (n)	29.4 (5/17) ^a	47.3 (9/19) ^a
Rejections (n)	70.5 (12/17) ^a	52.6 (10/19) ^a
Events		
Won (n)	35.3 (243/688) ^b	61.5 (539/876) ^a
Lost (n)	64.6 (445/688) ^a	38.4 (337/876) ^b
Horns		
PHo (n)	11.7 (2/17) ^a	36.8 (7/19) ^a
AHo (n)	88.2 (15/17) ^a	63.1 (12/19) ^a
Success index	0.34±0.0b	0.62±0.0a

^{a, b}Values within the same column, with different superscripts, differ ($P < 0.005$). Presence of horns (Pho) and absence of horns (AHo).

Table 2: Means (\pm SE) of the morphometric characteristics of Dor per rams of high (HR) or low (LR) social rank.

Rams (n)	LR (17)	HR (19)
HeL (M)	28.7±0.4 ^a	27.9±0.4 ^a
HeW (M)	10.1±0.6 ^a	10.2±0.2 ^a
HoL 1 (M)	6.1±1.0 ^a	7.4±0.9 ^a
HoL 2 (M)	10.5±0.8 ^a	7.5±0.6 ^a
HoW (M)	6.7±0.2 ^a	7.7±0.3 ^a
THoL (M)	8.3±0.9 ^a	8.0±0.6 ^a
Odor (M)	1.1±0.2 ^a	2.0±0.2 ^a

^{a, b}Values in the same column with different superscripts, are different ($P < 0.05$). Where: Head length (HeL), Head width (HeW), Horn length 1 (HoL 1), Horn length 2 (HoL 2), Horn width (HoW), Total horn length (THoL). M=Mean; n = Number.

Table 3: ASB, CSB and TEMP means of Dorper rams of high (HR) and low (LR) social ranks.

Rams (n)	LR (n = 17)	HR (n = 19)
Latency to ejaculation (s)	91.5±14.0 ^a	116.8±38 ^a
ASB (%)	0.7±0.1 ^a	0.3±0.5 ^b
Anogenital sniffing (M)	0.0±0.0 ^a	0.1±0.0 ^a
Approximation (M)	0.1±0.1 ^a	0.2±0.1 ^a
Kicking (M)	0.6±0.5 ^a	0.0±0.0 ^b
CSB (%)	2.3±1.0 ^b	3.3±1.1 ^a
Mount attempt (M)	0.3±0.2 ^b	0.7±0.2 ^a
Mount (M)	0.6±0.4 ^a	0.7±0.6 ^a
Mount with ejaculation (M)	1.3±0.5 ^a	1.4±0.4 ^a
TEMP (%)	0.3±0.3 ^a	0.3±0.1 ^a
Flight attempt (M)	0.3±0.0 ^a	0.2±0.0 ^a
Isolation (M)	0.0±0.0 ^a	0.1±0.1 ^a
Aggression (M)	0.0±0.0 ^a	0.1±0.0 ^a

^{a, b}Values with different superscripts between columns, differ ($P < 0.05$). The number of behavioral events considers 3 min during 5 consecutive days. Where ASB = Appetitive sexual behaviors; CSB = Consummatory sexual behaviors; TEMP = Stress temperament, M = Mean; N = Total number; S = Seconds.

not measured in the present study, further research would be necessary to ascertain this. In addition, by not having the competition of the dominant males, the low-ranking males were able to express their full sexual behavior. It is also possible that males of low hierarchy had to self-stimulate more to be able to achieve a mount with ejaculation, which would generate more ASB. Indeed, it has been shown that males with the presence of horns or with larger horns tend to be dominant (Hass and Jenni, 1991).

The dominant behavior that some males exert over

others of lower rank, will determine the reproductive success that each ram will achieve. In field conditions, low-rank rams decrease their frequency of mounting in the presence of a high-rank ram, but the latter do not modify their behavior in the presence of a low-rank ram (Lindsay *et al.*, 1976). In effect, high-ranking males had more CSB, even when low-ranking males had no competition when exposed to a female in heat. Furthermore, Tilbrook *et al.* (1987), found that in an extensive system the submissive rams, in the presence of the dominants, mount fewer females on fewer occasions

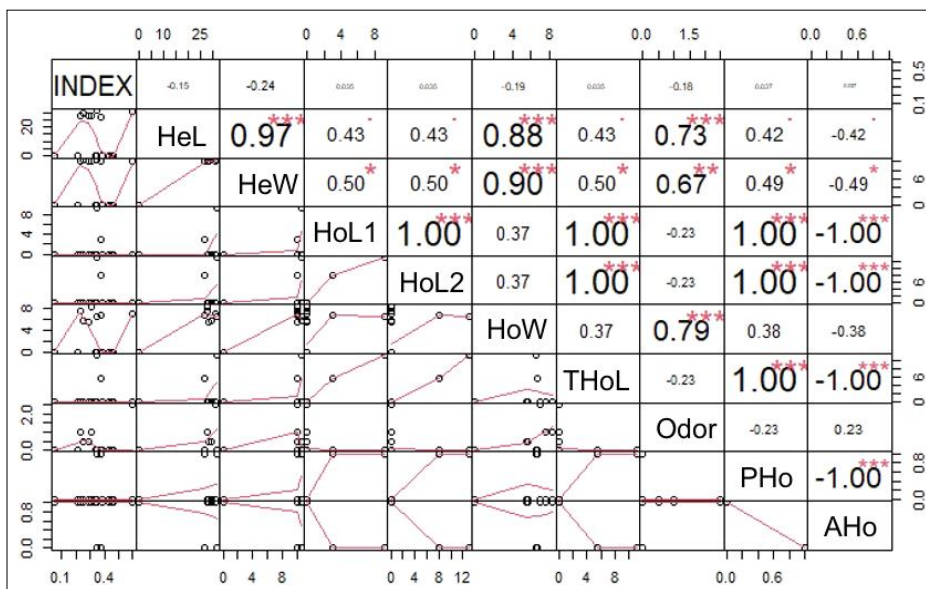


Fig 1: Spearman correlation for morphometric characteristics of low rank (LR) Dorper rams, where: Head length (HeL), Head width (HeW), Horn length 1 (HoL 1), Horn length 2 (HoL 2), Horn width (HoW), Total horn length (THoL), presence of horns (PHo) and absence of horns (AHo). *** Statistical difference at P<0.001, ** Statistical difference at P<0.01 and * Statistical difference at P<0.05.

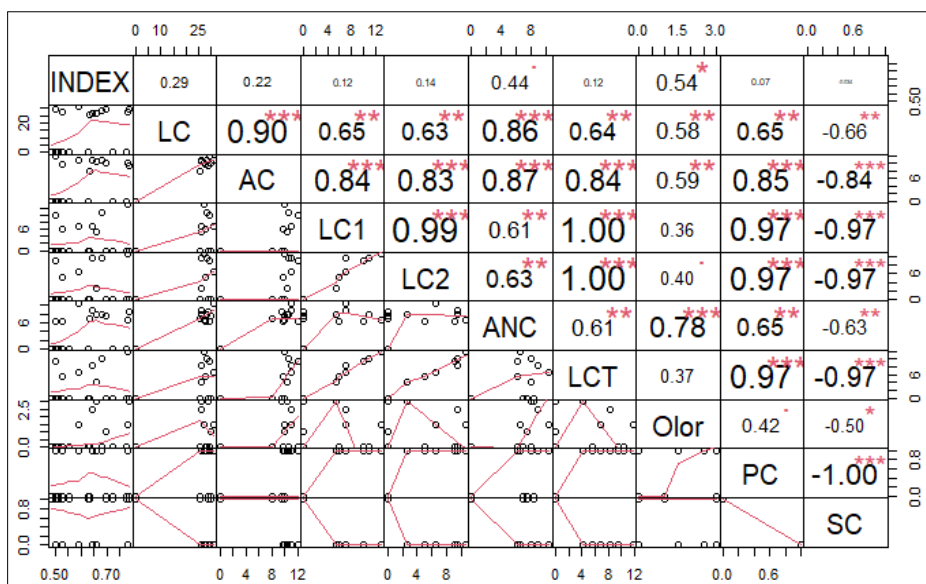


Fig 2: Spearman correlation for morphometric characteristics of high rank (HR) Dorper rams, where: Head length (HeL), Head width (HeW), Horn length 1 (HoL 1), Horn length 2 (HoL 2), Horn width (HoW), Total horn length (THoL), presence of horns (PHo) and absence of horns (AHo). *** Statistical difference at P<0.001, ** Statistical difference at P<0.01 and * Statistical difference at P<0.05.

and most copulations occur with the less preferred and attractive ewes for the dominants. However, our data are different from that reported by Ungerfeld and Nuñez, (2011), who suggest that high-ranking males have more capacity to perform more ASB and CSB than low-ranking males, attributed to the fact that high-ranking animals had better body development. They also mention that mid-ranking rams repeatedly show many mounts and mounts with ejaculation in the presence of low-ranking rams, but it is not observed in high-ranking rams when they are in the presence of mid- or low-ranking rams, when in contact with anestrus sheep (Ungerfeld, 2012). Another possibility for why the high-ranking males expressed more mounts and a shorter latency are some strategies of these males. For example, it has been shown that high-ranking males defend females in estrus but do not limit their movement (care), while subordinates try to abduct them from the dominants by blocking them (Hogg and Forbes, 1997). Another cause that could affect the CSB of males is stress since this can influence their social rank and it has been shown that animals more susceptible to stress are vulnerable to dominant males (Larrieu and Sandi, 2018). However, the results of this study suggest that males of either high or low hierarchy behaved similarly regarding stress. The measured behaviors of stress in this study were similar (TEMP), for which it's unlikely that they had any influence, even though it has been reported that males of the lower hierarchy are generally nervous because they sense the presence of males of higher hierarchy, even though they are not physically present (Ungerfeld and Nuñez, 2011).

CONCLUSION

Based on our results, we conclude that there are social hierarchies even when rams have the same morphometric characteristics regardless of social ranking, however, males of higher ranks express more consummatory sexual behaviors, while low social ranking males express more appetitive sexual behaviors when managed in intensive conditions.

Conflict of interest: None.

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