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Testicular biometry and hierarchy of dominance of the white-lipped peccary (Mammalia, Tayassuidae)

Biometria testicular e hierarquia de dominância do queixada (Mammalia, Tayassuidae)

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Abstract
The objective of this study was to obtain data on the testicular biometry of the white-lipped peccary (*Tayassu pecari*) and to verify if there is a correlation between the individual's biometric data with their dominance rank. Data were collected from 16 adult males, aged between two and seven years, who made up the same group. By the agonistic interactions analyses we calculated the linearity indexes (*h*') of the dominance hierarchy and dominance rank. At the end of the behavioral observations, males were captured to collect biometric data from the testis. White-lipped peccaries showed testicles located in a perineal position, inclined cranio-ventrally with oval shape, flattened laterally and with tenso-elastic consistency (2.54±0.07). There was variance in the means (± standard deviation) of testis length (5.88±1.05cm), width (4.24 ± 0.98cm), height (4.44±0.86cm), and total scrotal width (8.78±17.05). The linear dominance hierarchy described the social structure of the males of this group (*h*'>0.9), with probability of linearity in the hierarchy greater than chance (P = 0.02). There was a correlation between dominance rank with the body mass, length and volume of the testicles. As in general the dimensions of the testicles are directly correlated with sperm production and also testosterone, this characteristic favors the reproductive performance of dominant white-lips males. Therefore, the characteristics of testicular biometry of white-lips should be considered for the selection of individuals more likely to reproduce in captivity favoring the conservation of this vulnerable species.
Keywords: animal reproduction; applied ethology; captive breeding; endangered species; wildlife management

Resumo
O objetivo neste estudo foi obter dados sobre a biometria testicular do queixada (*Tayassu pecari*) e verificar se há correlação entre os dados biométricos do indivíduo com seu posto de dominância. Os dados foram coletados de 16 queixadas adultos, com idades entre dois e sete anos, que compunham o mesmo grupo. Por meio de análises das interações agonísticas foram calculados os índices de linearidade (*h'*) da hierarquia de dominância e determinado o *rank* dos indivíduos. Ao final das observações comportamentais, os machos foram capturados para coleta de dados biométricos do testículo. Os queixadas apresentaram testículos localizados em posição perineal, inclinados cranioventralmente com formato oval, achatados látero-lateralmente e com consistência tensoelástica (2,54±0,07). Houve variação nas médias (± desvio padrão) do comprimento testicular (5,88 ±1,05cm), largura (4,24±0,98cm), altura (4,44±0,86cm) e largura escrotal total (8,78±17,05). A hierarquia de dominância linear descreveu a estrutura social dos machos desse grupo (*h'*>0,9), com probabilidade de linearidade na hierarquia maior que o acaso (P = 0,02). Houve correlação entre a classificação de dominância com a massa corporal, comprimento e volume dos testículos. Como em geral as dimensões dos testículos estão diretamente correlacionadas com a produção de esperma e também de testosterona, essa característica favorece o desempenho reprodutivo de machos de queixadas brancos dominantes. Portanto, as características da biometria testicular de queixadas devem ser consideradas para a seleção de indivíduos com maior probabilidade de reprodução em cativeiro favorecendo a conservação desta espécie vulnerável.
Palavras-chave: animais silvestres; espécie ameaçada; etologia aplicada; manejo de fauna silvestre; reprodução animal

1. Introduction

In general, a positive correlation can be found between testicular size and testosterone production[(1)] and fertility.[(2)] In other words, the larger the testicles, the higher the amount of testicular parenchyma and the more competent the gonad will be to produce spermatozoa. On the other hand, small testicles have been correlated with infertility.[(3)] Based on this principle, the evaluation of

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testicular biometric parameters is a fundamental tool for predicting the reproductive potential and selection of males,[4,5] especially for species that live in promiscuous or polygamous systems in which different males can copulate with the same females during the same estrous period.[6]

Social dominance in several species is established through reproductive competition, and paternity is selected from individuals with the best aptitude for reproduction, with reproductive success, therefore, directly influenced by the dominance status.[7] Sexual selection operates when individual members of the more abundant same sex compete intensely for individuals who are members of the less abundant opposite sex, allowing the latter to choose individuals based on their traits.[8] This selection by females promotes competition and the evolution of reproductive traits in males.[8] Thus, it provides genetic variability and, on the other hand, reduces the probability of the birth of homozygous animals for lethal or deleterious recessive genes.[9]

White-lipped peccaries are social mammals that live in large groups formed by hundreds of individuals of both sexes and are an example of this selection.[10] The species has been undergoing the effects of excessive hunting and habitat loss and, therefore, is classified as vulnerable to extinction by the International Union for Conservation of Nature – IUCN.[11] Group life is known to offer a series of advantages, among which we can highlight collaboration in the search for food, higher ease in finding partners and reproducing, and help in defense against predators.[12,13] On the other hand, group life induces competition for resources. For this reason, some strategies in these species reduce the level of aggression and increase cohesion between individuals.[14]

In this sense, the dominance hierarchy is an effective strategy for recognizing leadership and avoiding agonistic encounters that could trigger serious injuries and fragmentation of the flock.[15,16] Moreover, the dominance hierarchy is advantageous because, once established, disputes are limited to ritualized confrontations, reducing the occurrence of disputes involving physical contact.[17] These ritualizations are a form of expression of aggressiveness through movements and/or demonstration of body parts to intimidate the opponent.[18]

White-lipped peccaries in captivity present a unique linear dominance hierarchy, which includes males and females under the leadership of a male, in most cases.[19,20] Furthermore, the dominant male may have priority over females in estrus. It may be because, in addition to being the heaviest,[19] they may also have larger testicles, which need to be checked. White-lipped peccary females are considered promiscuous[21] and these relationships are presumable to occur in the species: dominant males with larger testicular size. For this reason, dominant white-lipped peccary males are expected to have a larger testicular size and heavier body mass than submissive males. Assuming that animals in general have a positive correlation between testicular size and sperm production, the selection of animals with these traits could contribute to the reproduction and conservation of this endangered species.

2. Material and methods

2.1 Ethics note

The research was approved by the Animal Ethics Committee (CEUA) of the State University of Santa Cruz (UESC-BA), with protocol number 031/16.

2.2 Study site and experimental animals

Data were collected from 16 adult males born and raised in scientific captivity of wild animals at the State University of Santa Cruz (UESC), Ilhéus, BA, Brazil (legally registered by IBAMA, with process number 02006.000485/99-57). The animals were aged between two and seven years old, had an average weight of 37.8±4.8kg, and made up the same group along with six other adult females of the same origin. The group was housed in the same paddock with an earthen floor and delimited by a 1.5 m high wire fence supported by eucalyptus posts, covering an area of 940 m^2. The paddock had a trap corral to assist in the management, containment, and marking of the animals. It also had a brick masonry trough (0.6 m length x 0.3 m width x 0.2 m height) and three feeders made from recycled tires cut in half. The animals were fed twice a day, at 8:00 am and 4:00 pm, with a diet consisting of a mixture of corn, soybean meal, and mineral supplements, with water available ad libitum.

2.3 Collection of behavioral data

Before the beginning of the observations, the animals were contained with a containment net for individual marking with plastic earrings of different shapes to facilitate identification from a distance. Images of the animals were recorded by a digital camera (JVCGZ-HD500, Tokyo, Japan) positioned outside the paddock at a distance of 5.0 m from the feeders for 90 days, twice a day at feeding times (between 8:00 am and 9:00 am and between 4:00 pm and 5:00 pm), totaling 60 hours of observation. Subsequently, winners and losers of each agonistic social interaction were registered in an electronic spreadsheet after analyzing the interactions between them using the method of recording all occurrences[22] of agonistic interactions using the software Windows Media Player (Microsoft Corporation, USA, 2013). Only the agonistic interactions (aggression and submission) involving the males of the group were selected.

The frequency of occurrence of agonistic interactions per male and time of observation was calculated. These frequencies were compared using the non-parametric chi-square test. Subsequently, the data from agonistic interactions were analyzed to describe the social structure of white-lipped peccary males using the linearity index (*h'*) (Vries, 1998) [24], using the software SOCPROG 2.4.[23] The *h'* index ranges from zero to 1.0 and closer to 1.0 indicates higher linearity of the hierarchy. Additionally, SOCPROG 2.4 provided the significance level of the determined *h'* index value. Then, the ranks occupied by individuals in the dominance hierarchy (ranking) were determined using the I&SI method described by Vries,[24] which minimizes the effect of eventual ties (individuals with the same rank). Also, the directional consistency indices (DCI)[25] were determined for each dyad, and then the mean DCI of the group was calculated. DCI measures the direction of dominance within the hierarchy and ranges from zero (equal exchange of acts of aggression) to 1.0 (complete unidirectionality). The SOCPROG 2.4 program was also used to generate sociograms of agonistic interactions between males in the group.

2.4 Collection of testicular biometric data

The animals were transferred to individual pens at the end of behavioral data collection, two days before the beginning of collections, to prevent sexual activity from interfering with data collection. For the transfer, the males were contained using a net, weighed, and isolated in 12 m^2 pens surrounded by a wire mesh 1.5 m high. Importantly, the animals continued to receive the same diet and water ad libitum. On the day of collection, the animals were immobilized with the net, weighed, and submitted to the anesthetic protocol, which consisted of a combination of acepromazine and ketamine. First, acepromazine (Acepran 0.2%®, Univet S.A., São Paulo-SP, Brazil) was applied as preanesthetic medication at a dose of 0.2 mg/kg IM and, five minutes later, ketamine (Dopalen®, Vetbrands Saúde, Jacareí-SP, Brazil) was applied at a dose of 5 mg/kg IM, as described by Vieira et al.[26]

After drug application, the animals were monitored for desirable sedation, analgesia, and muscle relaxation to ensure a safe testicle measurement. For this purpose, the testicles were lightly pulled and positioned parallel within the scrotum. The shape, mobility, and consistency of the testicles were evaluated by palpation, varying on a scale from 1.0 to 5.0, as recommended by Unanian et al.[27] A caliper was used to measure the length of each testicle (craniocaudal measurement, excluding the head and tail of the epididymis), the width of each testicle (lateromedial measurement), the height of each testicle (dorsoventral measurement), and the total scrotal width (TSW) (lateral measurement of both testicles). The formula ($V = L \times W \times H \times 0.52$) was used to calculate the testicular volume (individual for each testicle), where L is the testicular length, W is the testicular width, and H is the testicular height.

2.5 Statistical analysis

The normality of testicular biometry data was verified by the Shapiro-Wilk test using the PROC UNIVARIANT procedure of SAS (9.3). Descriptive analyses were performed using the same procedure, in which the means and maximum and minimum values were determined, in addition to the coefficients of variation of each measure. Then, the symmetry between the testicles and the means of the biometric data were compared by univariate ANOVA using the PROC MIXED procedure of SAS (9.3). Subsequently, Pearson correlation analyses were performed using the PROC CORR procedure of SAS (9.3). The Spearman correlation test was used to verify the correlation between the ranking of males with their body mass and the biometric values of their testicles, using the software Stastistica (version 7.0, Stat Soft, Tulsa, OK, USA). A significance level of $P<0.05$ was considered for all analyses.

3. Results and discussion

In general, symmetry was observed between the right and left testicles in the studied animals (Table 1), corroborating the findings by Sonner et al.[28] White-lipped peccaries have testicles located in a perineal position, in an intermediate location between the anus and the inguinal region, more ventrally when compared to the testicles of pigs, with an oval shape and elongated cranioventrally, flattened latero-laterally similar to the testicles of collared peccaries, confirming the general descriptions by Sonner et al. and Dyce et al.[28,29] Additionally, white-lipped peccaries presented testicles with the presence of mobility within the scrotum and tensile elastic consistency, corroborating the findings for collared peccaries by Kahwage.[30]

Table 1. Means of testicular biometry parameters of white-lipped peccary (n=6).

Parameter (cm)	Right	Left	*P*	CV (%)*
Length	5.79	6.35	0.06	16.35
Width	4.39	4.39	0.98	18.54
Height	4.46	4.61	0.60	16.23
Volume	59.25	67.12	0.54	37.31
Consistency	2.55	2.53	0.65	3.03

*CV: Coefficient of variation.

White-lipped peccaries showed mean values for testicle length (5.88±1.05cm), width (4.24±0.98cm), height (4.44±0.86cm), and TSW (8.78±17.05) higher than the means described by Sonner et al.[28] It can be explained by the difference in the age of the animals in both studies. Sonner et al.[28] collected data from animals destined for slaughter, probably between 10 months and 1.5 years of age,[31] and, therefore, younger than those used in the present study. The tensile elastic consistency determined for white-lipped peccary in the present study is considered of intermediate value (2.54±0.07) and corresponds to the pattern found for pigs and is described as ideal for males in reproduction.[32]

In general, no asymmetry was found between the right and left testicles, only a slight superiority of the measured length and height (Table 1). This trait can be considered essential for the maintenance of good spermatogenic activity for breeding males considering that the physiological development of the gonads occurs equivalently, preserving the fusiform testicular shape and, therefore, maintaining the symmetry between them.[33] In fact, white-lipped peccary spermatozoa present high values of seminal viability, as described by Vieira,[34] who reported a mean value of 75.4±16.2% for the total motility.

A total of 349 agonistic interactions were recorded among the six males during the 60 hours of observation. No difference (X^2=1.23; P=0.54) was observed in the frequency of agonistic interactions (G1=11.6) per individual and time of observation in the studied group. The analysis of the agonistic interactions showed that the linear dominance hierarchy described the social structure in the white-lipped peccary group, with an *h'* index close to 1.0 and significant. The method to determine the *h'* employed in this study was described by De Vries (1998), which presupposes the principle of transitivity, in which if A dominates B and B dominates C, A dominates C even if no interactions are observed between A and C.[35]

The average DCI value between the dyads was 0.97±0.09, indicating stable asymmetric relationships – dominant animals in the ranking directing agonistic interactions to more subordinate animals. It reinforces the existence of a linear dominance hierarchy among white-lipped peccary males. In addition, a dominant animal in groups of white-lipped peccaries plays a fundamental role in maintaining group cohesion by constantly interfering with the agonistic interactions of others. A correlation was observed between the ranking and the body mass of the animals, with the heaviest animals occupying the highest ranks in the hierarchy (Table 2). It is in line with what was described by Nogueira-Filho et al.,[31] who reported that the heaviest animals are commonly positioned at the top of the social dominance hierarchy.

White-lipped peccaries inhabit tropical forests in groups that can reach hundreds of individuals.[10] Large groups composed of individuals of both sexes make the maintenance and cohesion of the group difficult. Thus, the existence of behavioral mechanisms, such as the social dominance hierarchy (mediated by visual, acoustic, and chemical signals), in which each individual recognizes its position within the flock, contributes to the reduction of conflicts before they become severe aggression, and may even result in the flock fragmentation. A correlation was observed between the ranking and testicle length and volume (Table 2). There was also a trend of correlations between ranking position and TSW (r_S= 0.77; P= 0.07), that is, dominant animals had larger size and testicular volume.

Table 2. Rank (α: dominant), body mass (kg), and codes of individuals in the three groups of white-lipped peccary

Code/ Rank*	Group 1 (Weight) kg	Code/ Rank	Group 2 (Weight) kg	Code/ Rank	Group 3 (Weight) kg
M1 (α)	42.0	M1 (α)	37.0	M1 (α)	35.0
M2	41.0	M2	37.2	M2	32.0
M3	41.0	M3	37.0	M3	34.5
M4	40.0	M4	38.3	M4	33.0
M5	33.0	M5	38.0	M5	30.3
M6	30.0	-	-	-	-

*Code/Rank: alphanumeric code of individuals in the groups (the letter identifies the sex, and the numbers indicate the rank in ascending order). α, animal considered dominant.

Table 3. Correlation coefficients between position (ranking) in the linear dominance hierarchy and testicular biometry parameters of males in G1 (N=6).

Parameter*	r_s
Right testicle	
Length	**0.94**
Width	0.60
Height	0.54
Volume	**0.83**
Testicular consistency	0.62
Left testicle	
Length	0.78
Width	0.71
Height	0.55
Volume	**0.83**
TSW	0.77
Testicular consistency	0.62

*Values in bold were statistically significant (P<0.05).

The ranking of white-lipped peccaries was correlated with body mass and testicle dimensions. More dominant animals had larger testicles than the most subordinate in the hierarchy. According to França[36] and Vieira,[37] there is a correlation between testicular size and sperm and hormone production, that is, the larger the testicles, the higher the amount of testicular parenchyma and the more competent the gonad is to produce sperm. In contrast, small testicles have been correlated with infertility.[3]

Heavier body mass and larger sizes of testicles may favor the reproductive performance of dominant males of groups of white-lipped peccaries, and the selection of animals with these traits certainly favors their reproduction in captivity. The results described in this study may help to optimize the reproductive management of white-lipped peccary, contributing to the conservation of this species vulnerable to extinction.

4. Conclusion

Male white-lipped peccaries are organized in a linear dominance hierarchy. The most dominant males in the hierarchy have a heavier body mass and larger testicles than subordinate animals. Heavier body mass together with higher testicular volume, as well as larger testicular length, may favor the reproductive performance of dominant males of white-lipped peccary groups. Therefore, the selection of animals with these traits may favor a system of rearing white-lipped peccaries in captivity by maximizing reproductive management, contributing to their conservation.

Declaration of conflict of interest

The authors declare that there is no conflict of interest.

Author contributions

Conceptualization: R.L.A. Vieira and M.A.R. Feliciano. *Data curation*: R.L.A. Vieira and C. H.S.C. Barros. *Investigation*: R.L.A. Vieira and M.A.R. Feliciano. *Methodology*: R.L.A. Vieira, D.E. Leonardo and A.K.S. Cavalcante. *Supervision*: R.L.A. Vieira and M.A.R. Feliciano. *Writing (original draft):* R.L.A. Vieira, M.C. Santos and P.M.O. Pedroso. *Writing (revision and edition)*: R.L.A. Vieira, M.C. Santos, P.M.O. Pedroso and A.K.S. Cavalcante.

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