

Assessment of the vertebral heart scale and cardiothoracic ratio to standardize the heart size of collared peccaries (*Tayassu tajacu* Linnaeus, 1758) restrained with ketamine and midazolam¹

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ABSTRACT.- Silva-Filho O.F, Pessoa G.T., Sousa F.C.A., Rodrigues R.P.S., Moura L.S., Ambrósio C.E., Silva A.B.S & Alves F.R. 2018. **Assessment of the vertebral heart scale and cardiothoracic ratio to standardize the heart size of collared peccaries (*Tayassu tajacu* Linnaeus, 1758) restrained with ketamine and midazolam.** *Pesquisa Veterinária Brasileira* 38(8):1705-1711. Departamento de Morfofisiologia Veterinária, Universidade Federal do Piauí, Campus Universitário Ministro Petrônio Portela, Bairro Ininga, Teresina, PI 64049-550, Brazil. E-mail: flavioribeiro@ufpi.edu.br

Wild species, especially those threatened with extinction, are increasingly being investigated to obtain information that can be useful for their preservation. The objective of the present study was to standardize the vertebral heart scale (VHS) and cardiothoracic ratio (CTR) of the collared peccary (*Tayassu tajacu* Linnaeus, 1758) sedated with ketamine and midazolam. Fourteen clinically healthy collared peccaries were examined in the two-year age group weighing 15-22kg. The animals were submitted to digital radiography of the thorax in lateral and dorsal ventral projections to calculate the VHS and CTR. The VHS mean values for males and females was 8.88 ± 0.51 v for right recumbency and 8.84 ± 0.39 v for left decubitus, and there were no significant between-gender differences regarding recumbency ($p > 0.05$). The CTR showed mean values of 0.50 ± 0.05 (males) and 0.45 ± 0.04 (females), but the gender-differences were not significant ($p > 0.05$). A positive correlation was shown between VHS and CTR ($r = 0.98$, right decubitus; $r = 0.96$, left decubitus). Establishing reference values for heart measurements in collared peccaries using digital radiography of the thorax permitted standardization of the VHS and CTR values for this wild species. In the studied wild animal model, the VHS and CTR heart assessment indexes were shown to be essential diagnostic tools for investigations of alterations in the size of the cardiac silhouette.

INDEX TERMS: Vertebral heart scale, cardiothoracic ratio, heart size, collared peccaries, *Tayassu tajacu*, ketamine, midazolam, cardiac silhouette, wild animals, cardiology, morphology.

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RESUMO.- [Avaliação do *vertebral heart scale* e índice cardiorácico na padronização do tamanho cardíaco de cateto (*Tayassu tajacu* Linnaeus, 1758) contidos com cetamina e midazolam.] Espécies silvestres têm sido cada vez mais exploradas como forma de obter informações que favoreçam sua conservação, especialmente aquelas ameaçadas de extinção. Este trabalho teve por objetivo a padronização dos valores de VHS (*vertebral heart scale*) e índice cardiorácico (ICT) de catetos (*Tayassu tajacu* Linnaeus, 1758) contidos com Cetamina e Midazolam. Foram avaliados 14 catetos

cl clinicamente saudáveis, com faixa etária de 2 anos e variação média de peso entre 15 a 22Kg. Os animais foram submetidos a radiografia digital de tórax em projeções laterolaterais e dorsoventrais para o cálculo do vertebral heart scale (VHS) e Índice Cardiotorácico (ICT). O VHS evidenciou valores médios entre machos e fêmeas de $8,88 \pm 0,51v$ para decúbito direito e $8,84 \pm 0,39v$ para decúbito esquerdo, não ocorrendo diferença estatística entre os decúbitos ($p > 0,05$). O ICT revelou valores médios de $0,50 \pm 0,05$ (machos) e de $0,45 \pm 0,04$ (fêmeas), não sendo verificada diferença estatística significativa entre os sexos ($p > 0,05$). Verificou-se correlação positiva entre VHS e o ICT ($r = 0,98$, decúbito direito, $r = 0,96$, decúbito esquerdo). O estabelecimento dos valores de referência para mensurações cardíacas em catetos, a partir de radiografias digitais do tórax, permitiu a padronização do VHS e ICT para esta espécie silvestre. No modelo animal silvestre estudado, os índices de avaliação cardíaca VHS e ICT mostraram-se como uma ferramenta diagnóstica imprescindível para investigações sobre as alterações do tamanho da silhueta cardíaca.

TERMOS DE INDEXAÇÃO: *Vertebral heart scale*, índice cardiotorácico, tamanho cardíaco, caititus, *Tayassu tajacu*, cetamina, midazolam, peccari, silhueta cardíaca, animais silvestres, cardiologia, morfologia.

INTRODUCTION

Radiographic examination is an image diagnosis method that is very important in veterinary medicine. The values obtained from VHS can help in the prediction of cardiac alterations that may reflect in clinical or surgical procedures (Carvalho 2004).

Thoracic radiographs are routinely used in human and animal medicine to assess alterations in cardiac silhouette conformation. In addition, this tool provides digital impressions of the appearance of the lung parenchyma, upper intrathoracic airways, anatomic integrity of the thoracic cavity walls, pleural space and lung vascularization (Lamb & Boswood 2002, Gabay 2003, Soares et al. 2004, Kealy & McAllister 2005).

Studies of the heart in radiographic images include the left and right decubitus positions; the dorsal ventral incidence (DV) is preferred to the ventral dorsal (VD) because the former better shows the appearance of the cardiac silhouette, avoiding outline distortions in the image. Furthermore, in the DV incidence, the caudal lung vessels present better radiographic definition, favoring diagnoses of lung congestion and permitting indirect assessment of heart performance (Ruehl & Thrall 1981, Lamb & Boswood 2002, Kealy & McAllister 2005).

The qualitative radiographic method is the one used most for heart assessments, where the veterinary or human radiologist relates anatomic knowledge to possible alterations in the radiographic image inside the thoracic cavity. The advantages of this method are convenience and speed, but it should be adopted only by experienced professionals who know the normal variations that the image can present (Suter & Lord 1971, Lamb & Boswood 2002, Soares et al. 2004).

Among the methods proposed, biometric measuring methods aim to assess heart size quantitatively to increase accuracy and decrease the subjectivity present in qualitative assessments (Buchanan & Bücheler 1995). In biometric methods, measurements of the heart dimensions and cardiothoracic relationships are necessary. In lateral projection, the heart length between the base and the apex should be approximately 70% of the dorsal ventral distance of the thoracic cavity (Schelling

2002), while the width or cranial-caudal diameter of the heart can range from 2.5 (in deep-chested dogs) to 3.5 intercostal spaces (in wide-chested dogs) (Buchanan & Bücheler 1995, Schelling 2002).

Studies on Brazilian fauna have high priority because of their ecological importance and the potential for animal science exploitation presented by various species (Neves et al. 2013). Basic information on wild animal morphology regarding their physiology and biology in captivity is lacking. Collared peccaries (*Tayassu tajacu*) are wild Suiformes belonging to the genus *Tayassu* and the family Dicotylidae. These animals are subject to intense predatory pressure, which has increased the need for greater knowledge regarding their cardiovascular physiology (Lazure et al. 2010, Azevedo et al. 2012).

The collared peccary is highly adaptable to different environments, extremely robust and resistant to various diseases. These characteristics greatly favor their use in scientific investigations, as observed by Bezerra et al. (2014) and Pessoa et al. (2014), which used these animals as models for renal ischemic lesions and the isolation of mesenchymal cells derived from adipose tissue. Thus, these animals are more accessible alternatives for scientific studies than is the domestic pig (*Sus scrofa domesticus*) (Monteiro et al. 2009) or mini pigs (Vodička et al. 2005).

Studies of heart measurements in domestic animals have been conducted for species such as dogs (Lamb et al. 2001), cats (Litster & Buchanan 2000), horses (Patteson et al. 1995), goats (Ukaha et al. 2013) and pigs. However, variables such as breed, animal size and thoracic shape have influenced the standardization of the mean vertebral scale of the normal heart, as has been observed in the domestic species already assessed (Azevedo et al. 2016).

Buchanan & Bücheler (1995) determined that values for dogs obtained in lateral projections were smaller or equal to 10.5 vertebra (v) in 98% of the radiography of clinically normal dogs. Corroborating the findings of these authors, Lamb et al. (2001) analyzed the use of specific VHS values as a method to diagnose heart disease in dogs. The authors reported that significant differences exist between the various mean values for different breeds, and suggested standardizing this technique for each breed in question.

The VHS (vertebral heart scale) is a well-established routine in veterinary medicine (Jepsen-Grant et al. 2013), and it correlates positively with cardiomegaly development in companion animals (Guglielmini et al. 2012). However, the VHS has been little used as a diagnostic tool in wild animals because standard values are that measure heart biometry available for only a few species, hindering the identification of determined abnormalities in wild animals. Felkai et al. (2014) claimed that the presence of pleural residual leakage and cardiomegaly were associated with increased VHS values in the primate *Cercopithecus neglectus*, which was later confirmed by echocardiographic examination. In humans, a cardiothoracic ratio of > 0.5 on a postero-anterior radiograph showed a good predictor of left ventricular dysfunction (Chana et al. 2015). Studies have aimed to standardize CTR values in nonhuman primates (Schillaci et al. 2009) and wild rodents (Moura et al. 2015), but are still scarce in several wild species, with no reports for species proposed in this research.

Rearing wild animals in captivity is a species management problem and requires safe and efficient restraint methods

that ensure adequate immobilization to carry out necessary procedures such as biological material collections, morphometric measurements, physiological measurements and clinical surgical interventions (Mayor et al. 2007). Thus, the objective of the present study was to standardize the VHS (vertebral heart scale) and cardiothoracic ratio (CTR) in collared peccaries (*Tayassu tajacu* Linnaeus, 1758) sedated with ketamine and midazolam.

MATERIALS AND METHODS

Animals. In this study, fourteen healthy adult collared peccaries (*Tayassu tajacu* Linnaeus, 1758) were used, seven males (20±1.19kg) and seven females (17.85±2.41kg), each approximately two years old, from the Nucleus for Wild Animal Study and Preservation (Núcleo de Estudos e Preservação de Animais Silvestres - NEPAS (Registro IBAMA nº 02/08-618) of the Agrarian Science Center (CCA) at the Federal University of Piauí (UFPI), Teresina, Piauí, Brazil).

The protocols used in the present study were approved by the Committee of Ethics in Animal Experimentation (Comitê de Ética em Experimentação Animal - CEEA/UFPI (nº 047/15) and authorized by the Ministry of the Environment through the Biodiversity Authorization and Information System (SISBIO) of the Brazilian Institute for the Environment and Renewable Natural Resources (IBAMA) (nº 48113-1).

Exclusion criteria. Subjects with an unremarkable clinical history and physical examination were included in the study. The cardiovascular and respiratory systems were inspected by auscultation of the heart valves and lung fields. The time of capillary perfusion and the diagnosis of dehydration were also evaluated by hemodynamic investigation. The systemic infectious processes were evaluated by palpating the peripheral lymph nodes (submandibular, cervical, and popliteal), abdominal palpation, and temperature measurement. In addition, all peccaries were subjected to blood, biochemical, and electrolyte tests. Animals with evidence of systemic diseases, cardiovascular abnormalities (murmurs or arrhythmias), or any degree of valve insufficiency observed on echocardiogram, or with excessive stress during the examination, were excluded from the study.

Anesthetic protocol. The peccaries were fasted (given no solid food) for 12 hours and unwatered for 6 hours and then captured in their enclosures with a capture net. For chemical sedation, a mixture of 15mg/kg ketamine hydrochloride (Vetanarcol®) with 3mg/kg midazolam maleate (Dormonid®) was administered intramuscularly. The procedures were started when the anesthetic effect was observed, 15 minutes after application.

Radiographic examinations. To conduct the radiographic examinations, the animals were placed in sternal and lateral (right/left) decubitus to obtain radiographic images in right and left dorsal ventral (DV) and lateral projections of the thoracic region. A mobile INTECAL CR-7 X-ray apparatus was used. The images acquired in a sensitive radiographic chassis were analyzed in a CR 30-X (Agfa HealthCare) digital radiographic image-capture apparatus installed in the Image Diagnosis Sector of the University Veterinary Hospital (HVU/UFPI), saved on a computer hard drive, and coupled to the analysis system.

VHS execution. Means were collected for the VHS (vertebral heart scale) according to the protocol established by Buchanan & Bücheler (1995) and Litster & Buchanan (2000), which directs sampling of the heart's length and width at its greatest diameter and subsequent comparison with the vertebral bodies from the cranial endplate of

the fourth thoracic vertebra (T4) (Fig.1). The measurements were taken first; later, the mean values were estimated to establish the normality criteria.

Cardiothoracic ratio. The CTR was assessed by comparing the greatest widths of the cardiac silhouette and the distance between the thoracic walls at height T8, according to methodology described by Schillaci et al. (2009) for *Macaca fascicularis*. Heart width was measured at its two greatest distances (C and D) from the vertical line that divides the limits between the right and left sides of the heart, at the point of its greatest diameter. Similarly, the thoracic width was measured at the point of greatest thoracic diameter (E) as follows:

$$CTR = \frac{C + D}{E}$$

Statistical analysis. The data were submitted to a normality test (Shapiro-Wilk and Kolmogorov-Smirnov test); then, group means were analyzed by Student's t-tests and paired for parameter interpretation at a confidence interval of 5% (p<0.05).

RESULTS

The animals assessed had cardiac silhouettes between the second and fourth intercostal spaces, situated on the mid-mediastinum in both decubitus (right lateral and left lateral) positions (Fig.1). The weight of the males studied averaged 20.0±1.19kg and the females averaged 17.85±2.41kg; there was no significant difference between the males and female weights (p>0.05), the mean weight of males and females combined was 19.06±2.04kg.

The VHS (vertebral heart scale) in the males showed a mean of 9.22±0.29v for the right decubitus and 8.87±0.42v for the left decubitus; there was no significant difference between them (p>0.05). For the females, the VHS showed a mean value of 8.55±0.47v for the right decubitus and 8.81±0.42v for the left decubitus; there was also no significant difference between them (p>0.05). When the VHS of males and females was compared for both of the decubitus positions, no difference was observed (p>0.05), and the estimated VHS was 8.85±0.39v (Table 1).

The cardiothoracic ratio (CTR) mean values were 0.50±0.05 for the males and 0.45±0.04 for the females, and there was no significant difference between the genders (p>0.05) (Table 1). Thus, the combined male and female CTR was 0.48±0.05. Positive correlation was observed between the VHS in both

Table 1. Comparative values measured for the vertebral heart scale and cardiothoracic ratio between the right and left lateral decubitus of collared peccary (*Tayassu tajacu* Linnaeus, 1758). Teresina, Piauí/Brazil, 2017

Parameters	Gender	Right decubitus	Left decubitus
VHS	Male	9.22±0.29	8,87±0,42
	Female	8.55±0.47	8.81±0.42
	Mean value to both decubitus	8.85±0,39	
CTR	Male	0.50±0.05	
	Female	0.45±0.04	
	Mean value	0.48±0.05	

VHS = vertebral heart scale, CTR = cardiothoracic ratio.

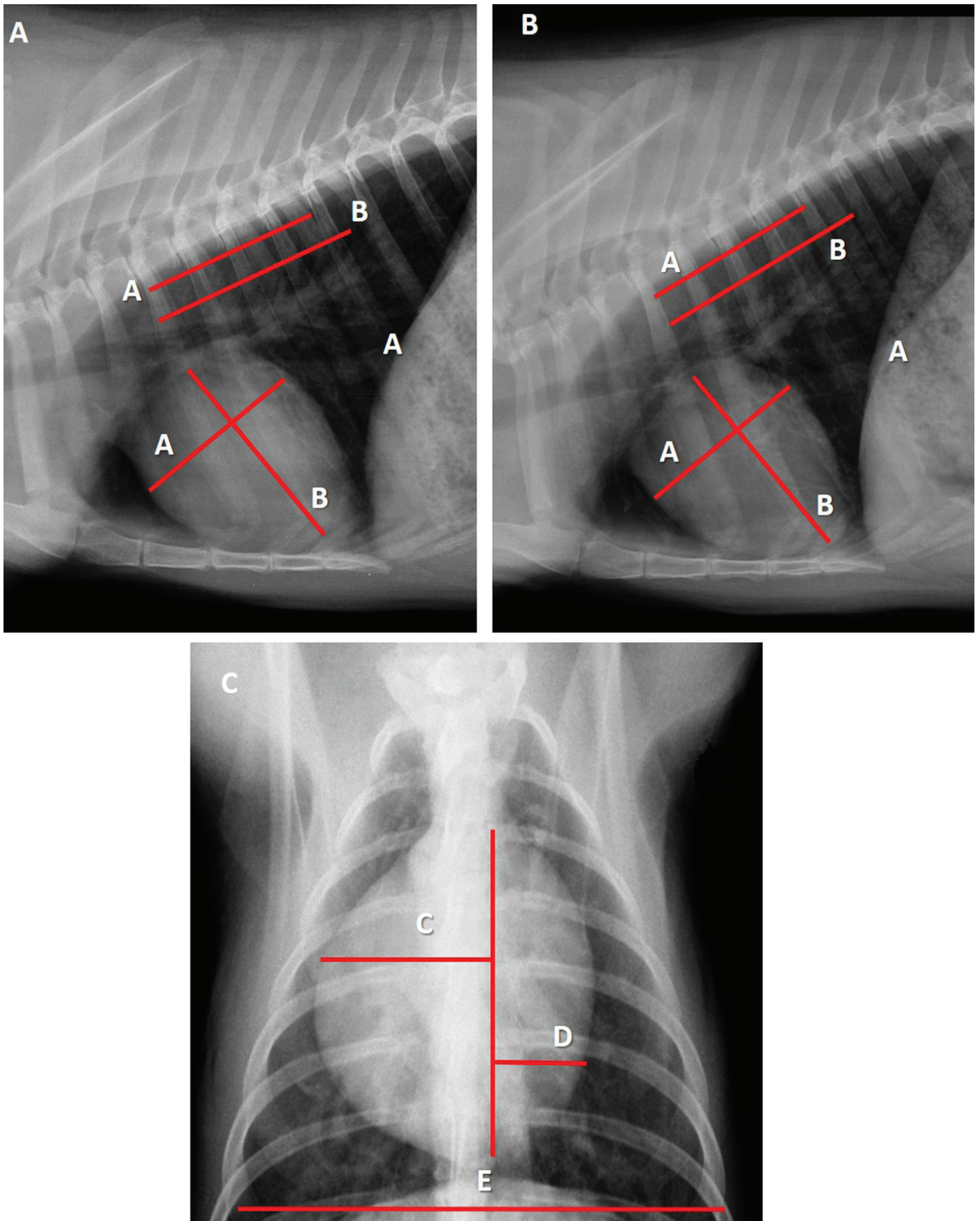


Fig.1. Digital radiographic image of the collared peccary thoracic cavity. (A) Right lateral projection. (B) Left lateral projection to obtain the vertebral heart scale (VHS) calculation. (C) Dorsal ventral projection to obtain the cardiothoracic ratio (CTR). Teresina, Piauí/Brazil, 2017.

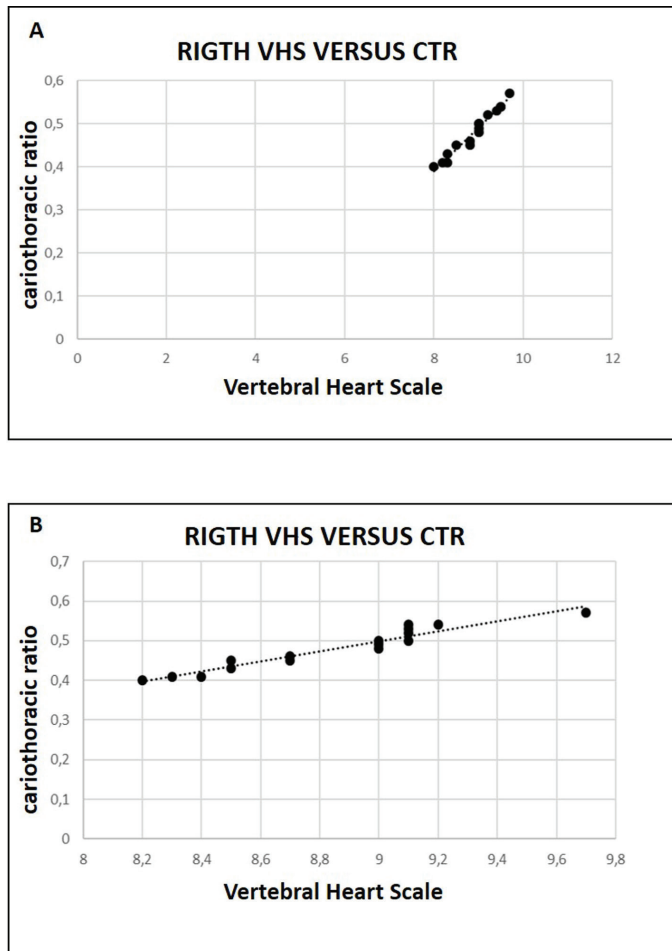


Fig.2. Correlation between the VHS and CTR of the peccary. (A) Linear correlation between vertebral heart scale (VHS) and cardiothoracic ratio (CTR) the in right lateral decubitus. (B) Linear correlation between VHS and CTR the in left lateral decubitus. Teresina, Piauí/Brazil, 2017.

decubitus and the CTR ($r=0.98$, right decubitus; $r=0.96$, left decubitus), taking CTR as the dependent variable (Fig.2).

DISCUSSION

The mean of the combined VHS values of collared peccary males and females ($8.88 \pm 0.51v$) was lower than that observed in other wild animals, such as coatis (*Nasua nasua*, $9.36 \pm 0.75v$; Martini et al. 2013), robust capuchin monkeys (*Cebus apella*, $9.25 \pm 0.32v$; Rocha-Neto et al. 2015), marmosets (*Callithrix jacchus*, $9.42 \pm 0.44v$; Wagner & Kirberger 2005) and alpacas (*Alpaca cria*, $9.36 \pm 0.65v$; Nelson et al. 2011); however, it was higher compared with ferrets (*Mustela putorius furo*, $5.39 \pm 0.45v$) and hedgehogs (*Atelerix albiventris*, $5.39 \pm 0.45v$) (Black et al. 2011).

There is a directly proportional correlation of the VHS (vertebral heart scale) values with the physical size of the animal, a fact that was observed when the collared peccary was compared with domestic Suiformes (*Sus scrofa domesticus*). The VHS values observed in the collared peccary were lower

than conventional pigs ($9.05 \pm 0.15v$) and close to the values found for mini pigs ($8.6 \pm 0.14v$).

Thoracic conformation affects the VHS values, a fact observed in narrow- and wide-chested dogs, as in the American Pit Bull Terrier breed, which presents lower and higher VHS values at 10.5 and 11.8 vertebrae, respectively (Cardoso et al. 2011). In contrast, in dogs with an intermediate-size thorax, such as the Indian Spitz, Labrador Retriever and crossbred dogs (SRD), the established VHS value is $9.7 \pm 0.5v$. Lastly, Bavegems et al. (2005) standardized the VHS variable at $11.0 \pm 0.5v$ (right lateral) and $11.3 \pm 0.5v$ (left lateral) for Whippets, dogs with a narrow and deep thorax, and significant differences were observed between the decubitus positions.

The results showed that the thoracic cavity morphology in collared peccaries varies little, similar to that observed in non-human primates, such as *Callithrix jacchus* (Wagner & Kirberger 2005), *Chlorocebus sabaues* (Young et al. 2013) and *Macaca mulata* (Ji et al. 2013). This characteristic is essential for applying biometric tests, enabling the detection of discrete increases in the cardiac silhouettes of these animals and permitting early diagnosis of cardiovascular diseases (Schillaci et al. 2010).

In the collared peccary, no significant statistical difference was observed between males and females for the CTR, a characteristic also observed in *tonkeana* and *cylopis* monkeys by Schillaci et al. (2009), and capuchin monkeys (*Cebus apella*) by Rocha-Neto et al. (2015) and in agoutis (*Dasyprocta prymnolopha*) by Moura et al. (2015). These results are probably due to behavioral characteristics in captivity, both for males and females; there were no marked differences in the cardio-hemodynamic profile in this wild species, and thus there was close conformation of the cardiac silhouette between the genders.

However, differences were observed in humans in CTR values between men and women (Giamouzis et al. 2008, Dimopoulos et al. 2013), and women presented a risk 2.5 to 4 times higher for developing left ventricular hypertrophy than did men (Chen et al. 2011).

The heart size ratios reflected in CTRs show a strong relationship with the health and youth of the animals. The collared peccaries studied were, at the physical examination, free from cardiorespiratory alterations and the lung fields were very audible. Studies of humans showed significant association between the CTR and malnutrition and anemia, afflictions that can lead to alterations in normal heart sounds (Chen et al. 2011). Further, the females were not pregnant during the collection periods, a fact that could result in alterations in the thoracic conformation of this portion of the experiment (Edelstein et al. 2005).

The CTR observed in the collared peccaries (0.50 ± 0.05) presented values like those observed in humans. Although we have not studied animals with the presence of cardiopathies, the proximity between these values suggests that, potentially, the peccary may present cut-off points close to those reported in humans for the determination of cardiac pathological alterations. Screatton (2010) demonstrated that the CTR was intimately related to the volume of the left ventricle. Rubens (1996) described CTR values of over 0.5 in humans as pathological. Hemingway et al. (1998) assessed the cardiothoracic ratio in adults with congenital cardiopathies and observed that patients with congenital heart diseases had a significantly

larger CTR (52.0%±7.6%) than did the control group formed by patients without alterations (42.3%±4.0%), and concluded that patients with high CTR presented a significantly higher risk of death. Furthermore, the CTR was a predictor of 13% of the mortality in patients with heart diseases, indicating the value of early monitoring and reduction of the CTR (Gao et al. 2009).

CONCLUSIONS

Establishing reference values for heart measurements in the collared peccary from digital radiography of the thorax permitted standardization of the vertebral heart scale (VHS) and cardiothoracic ratio (CTR) for this wild species.

The CTR showed positive correlation compared to the VHS, and can meet the needs of clinical precision in its application.

In the wild animal model studied, the VHS and CTR heart assessment indexes were shown to be essential diagnostic tools for studying alterations in the size of the cardiac silhouette.

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