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Radiographic pelvimetry in hybrid marmosets *Callithrix penicillata* (E. Geogfroyi, 1813) × *Callithrix jacchus* (Linnaeus, 1758)

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[Pelvimetria radiográfica em saguis híbridos Callithrix penicillata (E. Geogfroyi, 1813) × Callithrix jacchus (Linnaeus, 1758)]

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ABSTRACT

Radiographic pelvimetry is the measurement of pelvic diameters by radiographs. The results allow us to determine the pelvimetric pattern of a species, generating data for prophylactic methods against parturition complications and providing pelvis' anatomical classification. There is a lack of such data on wild animals. *Callithrix penicillata* and *C. jacchus* are two marmoset species introduced in Southeast Brazil, mainly by anthropic action, successfully adapted and hybridized. This study aimed to perform pelvic measurements through radiographic pelvimetry in hybrid marmosets (*Callithrix penicillata* × *Callithrix jacchus*) from different locations in the Rio de Janeiro state, Brazil. The radiographs were performed in the Veterinary Radiology Department of the Veterinary Hospital of UENF, undergoing measurements through the "Image J" program of upper (UBD), lower (LBD), middle (MBD) bi-iliac diameters, right (RDD), and left (LDD) diagonal diameter, sacropubic diameter (SPD), and pelvic inlet area (PIA). Analysis of variance was performed with different origin locations and sex of the specimens, and means were compared through the SNK Test at 5% probability. Pelvic diameters of females were significantly larger than males, with sexual dimorphism. The pelvis was classified as dolichopelvic. The obtained data can be pelvic reference values for these animals.

Keywords: Callitrichids, primates, pelvis, anatomy

RESUMO

Pelvimetria radiográfica é a obtenção de medidas dos diâmetros pélvicos por meio de radiografias. Resultados obtidos permitem determinar o padrão pelvimétrico de uma espécie, gerando subsídio para métodos profiláticos contra complicações de parto e proporcionando a classificação anatômica da pelve. Existe carência desses dados em animais silvestres. Callithrix penicillata e C. jacchus são duas espécies de saguis introduzidas no sudeste do Brasil, principalmente, por ação antrópica, as quais se adaptaram com sucesso e hibridizaram. Objetivou-se realizar mensurações pélvicas por meio da pelvimetria radiográfica em saguis híbridos (Callithrix penicillata X Callithrix jacchus) oriundos de diferentes localidades do estado do Rio de Janeiro, Brasil. As radiografias foram realizadas no setor de Radiologia Veterinária do Hospital Veterinário da UENF, passando por mensurações por meio do programa "Image J", de diâmetros biilíaco superior (DBS), inferior (DBI), médio (DBM), diâmetro diagonal direito (DDD) e esquerdo (DDE), diâmetro sacropúbico (DSP) e área de entrada da pelve (PIA). Foi realizada análise de variância, incluindo diferentes localidades de origem e sexo dos espécimes, e médias comparadas pelo teste de SNK a 5% de probabilidade. Diâmetros pélvicos das fêmeas foram significativamente maiores do que dos machos, havendo dimorfismo sexual. Classificou-se a pelve como dolicopélvica. Os dados obtidos permitem considerá-los valores de referências pélvicas para esses animais.

Palavras-chave: calitriquídeos, primatas, pelve, anatomia.

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INTRODUCTION

Neotropical primates of the genus *Callithrix* (Erxleben, 1777) are small animals, commonly called marmosets or monkeys, which comprise six species: *Callithrix jacchus, Callithrix penicillata, Callithrix aurita, Callithrix geoffroyi, Callithrix kuhlii,* and *Callithrix flaviceps* (Oliveira *et al.*, 2015).

Among the species of this genus, two of them are of real interest in this study, *Callithrix penicillata* and *C. jacchus*. They are known as black-tufted marmoset, native to the Central-West region of Brazil, and white-tufted marmoset, occurring in the Northeast region (Rylands, 1993; Oliveira *et al.*, 2015).

Hybridization between *Callithrix* sp. species has been reported since the 1970s. The hybridization of these two species in the state of Rio de Janeiro is not natural since both are exotic species for the region (Coimbra-Filho and Câmara, 1993). Invasive species can cause major impacts on species native to certain regions, threatening conservation. Hybrid marmosets are a potential threat to native callitrichid species of the Southeast region, such as the golden lion tamarin, since they have similar ecological aspects. There may be competition for food resources and refuge, in addition to the risk of introducing new pathogens, which may also be related to human diseases (Rylands, 1993).

Among the several species that are used in biomedical research, neotropical primates represent excellent experimental models, being an option for the destination of hybrid marmosets as models for the management of endangered species in captivity of the genus *Callithrix* and even to other callitrichids (Albuquerque, 2019). However, the information on basic and morphological research of marmosets is insufficient, mainly on different aspects of their anatomy (Bruni and Zimmerl, 1951).

Knowledge of the body's normal morphology and the proper structure designations are essential, both for learning and advancing all areas of medical science (Valle *et al...*, 2006), in addition to the growing effort to preserve and conserve wildlife. The scarcity of anatomy and parameters description in primates makes it difficult for veterinarians to work in this

segment, and one of the techniques for obtaining morphological descriptions in species is through radiographic pelvimetry (Ramadinha, 2003).

Given the importance of morphological knowledge of a species, the objective of this study was to describe the pelvic diameters of females and adult males of hybrid marmosets *Callithrix penicillata* × C. *jacchus* from two different locations in the state of Rio de Janeiro (RJ), the Área de Proteção Ambiental (APA) da Bacia do Rio São João/Mico-Leão-Dourado and Ilha d'Água. The results can serve as baseline data for a better understanding of the reproduction aspects of these hybrids, may assist in the reproduction management of other threatened callitrichids, and provide the pelvis' anatomical and obstetric classification.

MATERIALS AND METHODS

This study followed the ethical principles in animal experimentation adopted by the Brazilian Society for Laboratory Animal Science/Brazilian College of Animal Experimentation. It was approved by the Institutional Committee on Ethics of Animal Use (CEUA - UENF), Protocol No. 470/2021.

We used 10 carcasses of hybrid adult marmosets (Callithrix penicillata × Callithrix jacchus) listed by the Center for Teaching and Research in Wild Animals (Núcleo de Ensino e Pesquisa em Animais Selvagens - NEPAS) Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF), originating from the Bacia do Rio São João/Mico-Leão-Dourado. In addition, 22 radiographs of hybrid adult marmosets (C. $penicillata \times C. jacchus$) from the archive of the Sector of Studies in Ethology, Reintroduction and Conservation of Wild Animals (Setor de Estudos em Etologia. Reintrodução e Conservação de Animais Selvagens - SERCAS) and the Sector of Veterinary Radiology of the Veterinary Hospital of UENF, these animals were from removal in Ilha d'Água.

The radiographic study was carried out in the Veterinary Radiology sector from the Laboratory of Animal Morphology and Pathology (LMPA) of the Veterinary Hospital of UENF. The radiographic equipment was a conventional X-

ray, microprocessor, high frequency, 500 milliamperage (mAs) at 125 kilovolts (KVP) of the Philips brand.

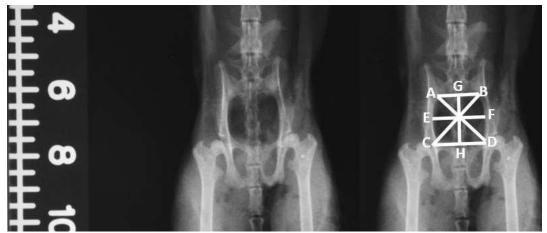
A 15 cm x 40 cm radiographic chassis was used, with a 15 cm x 40 cm Kodak TMAT G/RA radiographic film inside. For each radiographic image, a 30-centimeter (cm) scanning ruler was used on the chassis as a parameter method to correct projection differences. Each specimen was submitted to a radiographic image of a ventrodorsal (VD) projection.

The radiographic technique for VD projection was 160 mAs and 34 KVP with 0.045 seconds. The radiographs were developed in semi-automatic equipment of the Macrotec X-2 model, divided according to the group, and observed in a negatoscope. Subsequently, photographs were taken with a Nikon d 5100 professional camera to analyze the radiographic pelvimetry measures.

The radiographic images were evaluated and measured using the Image J software. Measuring

the upper bi-iliac diameter (UBD), lower bi-iliac diameter (LBD), middle bi-iliac diameter (MBD), right diagonal diameter (RDD), left diagonal diameter (LDD), and sacropubic diameter (SPD), according to the diameter methodology proposed by Ramadinha (2003).

The UBD was measured using a dashed line in a straight line transverse to the longitudinal diameter of the pelvis at the smallest diameter of the sacroiliac joint. Additionally, the LBD was measured through a straight line delineated parallel to the UBD at the iliopubic junction. The MBD measurement was performed parallel to the UBD and LBD through a transverse trace that extended from the left ilium bone to the right. The diagonal measurements, RDD and LDD, were performed with intersection points at the smallest diameter of the sacroiliac joint and the iliopubic junction. The SPD was obtained from the sacral promontory to the cranial part of the pubic symphysis, as shown in Figure 1.



Source: Laboratory of Animal Morphology and Pathology (UENF), 2020.

Figure 11. Scanned radiograph of hybrid marmoset bony pelvis ($Callithrix\ penicillata \times Callithrix\ jacchus$) with definitions of the measured diameters: AB - upper bi-iliac diameter; CD - lower bi-iliac diameter; EF - middle bi-iliac diameter; AD - right diagonal diameter; BC - left diagonal diameter; GH - sacropubic diameter.

According to the methodology proposed by Oliveira *et al.* (2003), the pelvic inlet area (PIA) was also measured, calculated by the Microsoft Excel program through the formula: PIA=(MBD) X (SPD) X π 2. The obtained data were tabulated in centimeters for each specimen for all measured variables.

The radiographic images were divided into a large group of male specimens from the APA and Ilha d'Água and female specimens from both locations. Depending on sex and location, the marmoset radiographs were classified into four other groups, namely: GI (males from APA), GII

(females from APA), GIII (males from Ilha d'Água), GIV (females from Ilha d'Água).

In addition, another division formed a group 'R', composed of all radiographs of males and females from the APA, and group 'D', composed of all radiographs of males and females from Ilha d'Água. Accordingly, the normality test (univariate PROC) was performed for all variables. Subsequently, the analysis of variance was performed through the GLM procedure from

SAS software (2019) with effects of group and location. The means between the groups were compared by the Student-NewmanKeuls test (SNK) at a 5% probability.

RESULTS

For the 32 specimens, the means and standard deviations for radiographic pelvimetry are presented in Tab. 1, 2, and 3.

Table 1. Means and standard deviations in centimeters for pelvic inlet area (PIA), upper bi-iliac diameter (UBD), lower bi-iliac diameter (LBD), middle bi-iliac diameter (MBD), right diagonal diameter (RDD), left diagonal diameter (LDD), and sacropubic diameter (SPD) of hybrid marmosets (*Callithrix penicillata* × *Callithrix jacchus*) in different groups. M = males from the Área de Proteção Ambiental (APA) da Bacia do Rio São João/Mico-Leão-Dourado and Ilha d'Água - Baía de Guanabara, RJ, F = females from APA and Ilha d'Água - Baía de Guanabara, RJ

Al A and fina d'Agua - Data de Guanabara, Ki					
	Sex				
Variable	M	F			
PIA	1.91 ± 0.33 B	$2.59\pm0.28~^{\mathrm{A}}$			
UBD	1.21 ± 0.11^{B}	$1.48\pm0.12~^{\mathrm{A}}$			
LBD	1.28 ± 0.15 B	1.53 ± 0.09 ^A			
MBD	$1.37\pm0.09~^{\rm B}$	$1.62 \pm 0.07^{\text{ A}}$			
RDD	1.76 ± 0.17 B	$2.07\pm0.10^{\rm \ A}$			
LDD	1.77 ± 0.15 $^{\rm B}$	$2.07\pm0.15~^{\mathrm{A}}$			
SPD	$1.76\pm0.26~^{\mathrm{B}}$	2.03 ± 0.15 ^A			

Means followed by the same letters in a row do not differ from each other by the SNK test at a 5% probability.

Table 2. Means and standard deviations in centimeters for pelvic inlet area (PIA), upper bi-iliac diameter (UBD), lower bi-iliac diameter (LBD), middle bi-iliac diameter (MBD), right diagonal diameter (RDD), left diagonal diameter (LDD), and sacropubic diameter (SPD) of hybrid marmosets (*Callithrix penicillata* × *Callithrix jacchus*) in different groups: GI = males from the Área de Proteção Ambiental (APA) da Bacia do Rio São João/Mico-Leão-Dourado; GII = females from APA; GIII = males from Ilha d'Água - Baía de Guanabara, RJ; and GIV = females from Ilha d'Água - Baía de Guanabara, RJ

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	Group			
Variable	GI	GII	GIII	GIV
PIA (cm)	$2.26\pm0.38~^{\rm A}$	2.55 ± 0.27 $^{\rm A}$	$1.75 \pm 0.16^{\text{ B}}$	2.61 ± 0.29 ^A
UBD (cm)	1.28 ± 0.83 ^C	$1.58 \pm 0.10^{\text{ A}}$	1.19 ± 0.11 ^C	1.43 ± 0.11^{B}
LBD (cm)	$1.30 \pm 0.15^{\ B}$	$1.52 \pm 0.10^{\text{ A}}$	$1.28\pm0.15~^{\rm B}$	1.54 ± 0.09 ^A
MBD (cm)	1.46 ± 0.54 B	1.64 ± 0.11 ^A	1.33 ± 0.08 ^C	$1,61 \pm 0.06$ ^A
RDD (cm)	$1.80 \pm 0.20^{\ B}$	2.14 ± 0.15 $^{\rm A}$	$1.75\pm0.16^{\ B}$	$2.04\pm0.05^{\mathrm{A}}$
LDD (cm)	$1.80 \pm 0.20^{\ \text{C}}$	2.20 ± 0.14 $^{\rm A}$	$1.76 \pm 0.13^{\text{ C}}$	2.01 ± 0.12^{B}
SPD (cm)	1.98 ± 0.38 ^A	$1.98 \pm 0.10^{\text{ A}}$	$1.67 \pm 0.10^{\ B}$	$2.05 \pm 0.17^{\text{ A}}$

Means followed by the same letters in a row do not differ from each other by the SNK test at a 5% probability.

Table 3. Means and standard deviations in centimeters for pelvic inlet area (PIA), upper bi-iliac diameter (UBD), lower bi-iliac diameter (LBD), middle bi-iliac diameter (MBD), right diagonal diameter (RDD), left diagonal diameter (LDD), and sacropubic diameter (SPD) of hybrid marmosets (Callithrix penicillata×Callithrix jacchus) from different locations: R=specimens from the Área de Proteção Ambiental da Bacia do Rio São João/Mico-Leão-Dourado; D=specimens from Ilha d'Água – Baía de Guanabara, RJ

	Location		
Variable	R	D	
PIA	2.40 ± 0.35 ^A	$2.18 \pm 0.49^{\text{ B}}$	
UBD	$1.43 \pm 0.18^{\text{ A}}$	$1.31 \pm 0.16^{\text{ B}}$	
LBD	$1.41 \pm 0.17^{\text{ A}}$	1.41 ± 0.18 ^A	
MBD	$1.55 \pm 0.12^{\text{ A}}$	$1.47 \pm 0.16^{\text{ B}}$	
RDD	1.97 ± 0.24 ^A	1.90 ± 0.19 ^A	
LDD	2.00 ± 0.26 ^A	1.89 ± 0.18 ^A	
SPD	1.98 ± 0.26 ^A	1.86 ± 0.24 ^A	

Means followed by the same letters in a row do not differ from each other by the SNK test at a 5% probability.

DISCUSSION

Determining the means and standard deviations of pelvic diameters not only contributes to the observation of sexual dimorphism but also helps as a prophylactic method of obstetric dystocia of different species. In addition, it delimitates planes and diameters in the several segments of the pelvis, allowing the study and fetal location with greater precision (Eneroth; Haughey, 1999) since species with small bodies and developed brains, such as primates, may have greater difficulty in parturition due to cephalopelvic disproportion (Trevathan, 2015).

In the evaluation of pelvic diameters of hybrid marmosets of this study, it was observed that all females from the different locations (F) presented significantly higher average pelvic dimensions than males (M). These results show the sexual dimorphism of these animals through the technique, as already described for the common squirrel monkey (Saimiri sciureus), Colombian red howler (Alouatta seniculus), lar gibbon (Hylobates lar), black lion tamarin (Leontopithecus chrysopygus), golden lion tamarin (L. rosalia), golden-headed lion tamarin (L. chrysomelas) (Ramadinha, 2003), and whitetufted marmoset (Callithrix jacchus) (Pinheiro et al., 2016).

When analyzing the means for the pelvic inlet area (PIA) of males from the APA (GI), and the two groups of females (GII; GIV), no significant statistical difference was observed between them. Schultz (1949) states that differences in PIA are better observed in large apes such as orangutans

(*Pongo pygmaeus*) and gorillas (*Gorilla gorilla*) than in small monkeys.

However, considering the whole group of females (F) and the whole group of males (M), there is a significant difference for the PIA, a similar result to that described for Azara's night monkey (*Aotus azarae*) (Valle *et al.*, 2006) and white-tufted marmoset (*Callithrix jacchus*) (Pinheiro *et al.*, 2016), being justified by an adaptation of the birth canal in these animals (Favoretto *et al.*, 2018).

A study by Aksel and Abee (1983) highlights the importance of measuring this area. The authors report a significant difference between two groups of common squirrel monkeys (*Saimiri sciureus*), in which the PIA was analyzed between females that had normal parturition (1.81cm) and females that gave birth to dead fetuses (1.64cm), demonstrating that obstetric complications can be directly related to low PIA values.

Favoretto *et al.* (2018) show that common squirrel monkeys (*Saimiri sciureus*) have a high rate of obstetric complications in the parturition of captive animals. Something similar is observed in golden-headed lion tamarins (L. *chrysomelas*), where the high number of dystocic births contributes to the species' risk of extinction (Ramadinha, 2003).

Therefore, radiographic pelvimetry can be considered an evaluation tool for reproduction management and species conservation programs, assisting in selecting and removing animals from

the reproduction colony that presents narrowing of the pelvis and preventing surgical interventions (Valle *et al.*, 2006).

Considering the upper bi-iliac diameter (UBD), although there is a significant difference in the mean between females from different locations (GII; GIV), they presented higher values than males (GI; GIII), as well as for the lower bi-iliac diameter (LBD). The LDB is directly linked to parturition, in which the sacropubic diameter is the one that varies the least according to the number of births that a female may have, concluding that the metric modifications that occur in the pelvis can be proportionally related to the number of births, and arise more from the increase in its width (LDB) than its height (SPD) (Guerrero *et al.*, 2016).

Similar findings regarding the significant difference of these variables between females and males were described for Azara's night monkey (*Aotus azarae*) (Valle *et al.*, 2006) and white-tufted marmoset (*Callithrix jacchus*) (Pinheiro *et al.*, 2016). However, the results differ from those described for black lion tamarin (L. *chrysopygus*), in which males have average UBD and LBD higher than females of the same species. Thus, significant differences exist between the bony pelvis of neotropical primates (Ramadinha, 2003).

Regarding the middle bi-iliac diameter (MBD), females from APA and Ilha d'Água (GII; GVI) presented higher values than males from both locations (GI; GIII), similar to what is described for lion tamarins (*Leontopithecus* sp.) (Ramadinha, 2003), Azara's night monkeys (*Aotus azarae*) (Valle *et al.*, 2006), and white-tufted marmosets (*Callithrix jacchus*) (Pinheiro *et al.*, 2016). Li (2002) points out that knowledge of this variable is essential for measuring PIA.

Regarding the right diagonal diameter (RDD) and left diagonal diameter (LDD), we found lower measures for males of different locations (GI; GIII) than females (GII; GIV), similar to that reported for white-tufted marmosets (Callithrix jacchus) (Pinheiro et al., 2016) and Azara's night monkeys (Aotus azarae) (Valle et al., 2006). However, females from APA (GII) had a higher LDD than those from Ilha d'Água (IV). Observing mean values between RDD and LDD of APA males (GI), it is noted that the

dimensions were equal, which resembles the study of Ramadinha (2003). However, for all other groups (GII; GIII; GIV), the values between RDD and LDD differ, diverging from the last author and corroborating with Pinheiro *et al.* (2016).

Regarding the sacropubic diameter (SPD), there was no significant difference between males and females from APA (GI; GII) and females from Ilha d'Água (GIV), according to what is described for lion tamarins (*Leontopithecus sp*) (Ramadinha, 2003), Azara's night monkeys (*Aotus azarae*) (Valle *et al.*, 2006), and white-tufted marmosets (*Callithrix jacchus*) (Pinheiro *et al.*, 2016). However, analyzing only males and females from Ilha d'Água (GIII; GIV), the results are similar to those described by these authors above, i.e., there is a significant difference between males and females, emphasizing the singularity for each species and the importance of determining anatomical parameters.

Comparing the whole marmoset group from APA (R) with those from Ilha d'Água (D), only the variables PIA, UBD, and MBD were significantly different, in which marmosets from APA (R) had higher means than the others. In lion tamarins, there are significant differences in MBD, LBD, and SPD between animals from free or captive life (Ramadinha, 2003). Albuquerque (2019) points out that captive primates may receive inadequate diets, especially regarding calcium concentrations, since the diet is usually composed of fruits, which do not have a large amount of this mineral.

Marmosets from Ilha d'Água had more contact with humans, and primates tend to share their food in this situation. Also, the intake of foods rich in fats and sugars may occur, leading to metabolic disorders and consequent mineral deficiency (Albuquerque, 2019). Siragusi (2019) describes cases of narrowing of the pelvis associated with vitamin D deficiency in five black-tufted marmosets (Callithrix penicillata), four females and one male, and it should be noted that they were all animals from the illegal wildlife trade. Primates kept as pets may develop vitamin D deficiency, and even if this can be treated with dietary therapy, some animals will remain with bone and 46 joint deformities, which may induce limitations (Johnson-Delaney, 1994).

Although animals from Ilha d'Água had some lower pelvic diameters than those from APA, bone demineralization was not observed in the radiographic study since this sign is considered one of the most important clinical signs of vitamin D deficiency (Albuquerque, 2019).

Regarding the classification of the pelvis type of these hybrid marmosets (*Callithrix penicillata* × *Callithrix jacchus*), according to the data obtained in the present study, it can be said that they are dolichopelvic since the MBD values are lower than SPD for both sexes and regions, similar to that described for lion tamarins (Ramadinha, 2003), Azara's night monkeys (Valle *et al.*, 2006) white-tufted marmosets (Pinheiro *et al.*, 2016), and ruminants (Oliveira *et al.*, 2003).

CONCLUSIONS

There is a significant difference in pelvic diameters between males and females of hybrid marmosets *Callithrix penicillata* × *Callithrix jacchus*, confirming sexual dimorphism by the technique of radiographic pelvimetry.

Hybrid marmosets from the Área de Proteção Ambiental da Bacia do Rio São João/Mico-Leão-Dourado have higher pelvis inlet area, upper and middle bi-iliac diameter than those from Ilha d'Água.

Morphologically, this study classifies hybrid marmosets as having a dolichopelvic bony pelvis.

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