

ORIGINAL ARTICLE

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# Sexual dimorphism in the whole brain and brainstem morphometry in the African giant pouched rat (*Cricetomys gambianus*, Waterhouse 1840)

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The study was conducted to evaluate the sexual dimorphism in the morphometric parameters of the whole brain and the brainstem in adult captive African giant pouched rats. Twenty-nine brain samples, comprising 15 females and 14 males, were utilized. Following brain extraction by standard procedures, the mean values of the weights, lengths, diameters, and volumes of the intact brains and the brainstem structures were compared in male and female rats using quantitative analytical statistical methods. The absolute brain weight obtained in the male was significantly higher (p < 0.01) than that of the female while the relative brain weight obtained in the female (0.636  $\pm$  0.049%) was higher than that of the male rats (0.564  $\pm$  0.032%), although the difference in the values was not statistically significant (p > 0.05). The lengths of the brain and myelencephalon, as well as the weights of the myelencephalon and mesencephalon, did not differ between the two sexes (p > 0.05). The weights and lengths of the pons did not differ between the two sexes (p > 0.05). There was a significant difference (p < 0.05) in the pyramidal length and a highly significant difference (p < 0.01) in the pyramidal width between the two sexes, with the male rats having higher values. This pioneering sexual dimorphic brain morphometry provides information for further research. (Folia Morphol 2010; 69, 2: 69–74)

Key words: mesencephalon, pons, myelencephalon, sexual differences

### INTRODUCTION

The African giant pouched rat (*Cricetomys gambianus*) is a representative of the *Muridae* family, which constitutes a group of the order *Rodentia*. They are also known as "fancy rats" or "comic rats" due to their comical facial expression whilst they have an engorged pouch. They are found in Central and West African countries, including Nigeria [10]. This wild rodent is currently domesticated and bred, hence the need to understand their specific biology.

The brainstem is made up of the myelencephalon, pons, and mesencephalon [22, 23]. Some authors [1, 9] are of the view that the diencephalon is part of the brainstem because it is a rostral continuation of the mesencephalon, and it retains the tubular structure of the neural tube. However, from an ontogenetic perspective, the diencephalon and telencephalon are of the same origin: the prosencephalon. Also, the diencephalon is functionally closely related to the telencephalon. Thus, we con-

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sidered the diencephalon to be a non-component of the brainstem.

Morphometric analysis of organs is essential. This is because such analysis may expose small structural changes that cannot be observed by ordinary qualitative analysis. There is a dearth of information on the sexual differences in the whole brain and brainstem morphometry for the African giant pouched rat in the available literature. Consequently, the aim of the study was to obtain morphometric data on the whole brain and brainstem of the African giant pouched rat with respect to sex so as to offer a reference for future research on comparative neuroanatomy of the rodent.

## **MATERIAL AND METHODS**

#### Experimental animals and management

Twenty-nine captive and clinically healthy adult African giant pouched rats were used for this study. The animals were captured live from the wild in Zaria, Kaduna State, Nigeria, using locally made traps. They were transported by road in standard laboratory cages to the animal pen of the Department of Veterinary Anatomy, Ahmadu Bello University, Zaria, where they were acclimatized for one month before the commencement of the experiment. The animals were given access to food and drinking water *ad libitum* throughout the experimental period.

#### Brain extraction and morphometric assessment

The body weight of each rat was obtained with a digital electronic balance [Citizen Scales (1) PVT Ltd., sensitivity: 0.01 g]. The rats were then euthanized by an overdose of inhalation chloroform anaesthesia. Each skull, containing the brain, was obtained after skinning and stripping off all the facial muscles within 30 minutes post-slaughter. Immediately, craniotomy through the calvaria revealed the dura mater which was cut with a pair of curved pointed scissors. The falx cerebri and tentorium cerebelli were both pulled from the longitudinal and transverse fissures of the brain, respectively, by gentle traction. At this stage, the brain (still within the cranium) was fixed in 10% phosphate-buffered formalin for 3 days to facilitate its easy extraction according to the method of Ramaswamy [19]. The extracted brains were examined and found to be devoid of any pathology. They were then weighed using a sensitive electronic balance (Mettler balance P 1210, Mettler instruments AG. Switzerland; sensitivity: 0.001 g). Brain volumes were estimated by the weight displacement method as described by Scherle [21]. Brain lengths were obtained using a vernier calliper (MG6001DC, General Tools and Instruments Company, New York; sensitivity: 0.01 mm).

In order to remove the brainstem from the rest of the brain, the cerebrum was firstly separated thus: the two cerebral hemispheres were gently pulled apart at the occipital pole to expose the corpus callosum. The entire corpus callosum together with the septum pellucidum and the body and rostral commissure of the fornix were severed in the midline. The diencephalon was severed at the level of the 3<sup>rd</sup> ventricle. This completely separated the forebrain from the brainstem and cerebellum. In order to separate the cerebellum from the brainstem, the floculi of the cerebellum were manually raised to expose the cerebellar peduncles. These peduncles were then severed on both sides. Finally, the brainstem was freed from the *arachnoidea* and cranial nerves by simple trimming. Using a scalpel blade, the mesencephalon was separated from the pons, and an incision at the pontomedullary junction separated the myelencephalon from the pons. The dimensions and volumes of the brainstem structures were obtained using the same instruments mentioned above. Landmarks for the measurement of dimensions of the brainstem structures were as follows: Brain length: rostrocaudal extent from tip of olfactory bulb to pyramidal decussation, at the level of the foramen magnum. Length of mesencephalon: rostrocaudal extent of the lateral surface of the corpora quadrigemina. Width of mesencephalon: transverse extent of the caudal surface of the mesencephalon, rostral to the pons. Diameter of colliculus: transverse extent of one colliculus. Height of colliculus: dorsoventral extent of one colliculus. Length of pons: extent of the pons along the width of the brain. Width of pons: extent of the pons along the rostrocaudal axis of the brain. Length of myelencephalon: rostrocaudal extent from the pontomedullary junction to the pyramidal decussation. Length of pyramid: rostrocaudal extent of one pyramid. Width of pyramid: transverse extent of one pyramid.

All recorded weights, lengths, widths, and volumes of the whole brain and brainstem structures were expressed as mean ( $\pm$  standard error of the mean) and subjected to statistical analysis. Student's t-test was used to determine the significance of the difference in the values obtained in male and female animals. Values of p < 0.05 were considered significant.

#### RESULTS

The mean body weight and nose-rump length obtained from all the 29 rats, irrespective of their sex,

Brainstem structure	Minimum	Maximum	Mean ± SEM
Mesencephalon weight [g]	0.300	0.800	0.545 ± 0.241
Mesencephalon length [mm]	3.530	7.830	$4.885 \pm 0.198$
Mesencephalon width [mm]	1.050	9.300	$6.886\pm0.361$
Rostral colliculus diameter [mm]	0.700	1.900	$1.396 \pm 0.068$
Rostral colliculus height [mm]	3.050	6.060	$4.421 \pm 0.144$
Caudal colliculus diameter [mm]	2.000	3.610	$2.515 \pm 0.080$
Caudal colliculus height [mm]	5.010	8.050	$6.943 \pm 0.162$
Pons weight [g]	0.100	0.270	0.171 ± 0.007
Pons length [mm]	2.280	4.920	$3.436 \pm 0.125$
Pons width [mm]	1.300	1.900	$1.686 \pm 0.028$
Myelencephalon weight [g]	0.410	0.800	$0.570 \pm 0.018$
Myelencephalon length [mm]	6.970	17.080	$10.200 \pm 0.327$
Myelencephalon volume [mL]	0.500	0.700	$0.583 \pm 0.012$
Pyramid length [mm]	0.730	10.170	$7.950 \pm 0.339$
Pyramid width [mm]	0.830	2.080	$1.482\pm0.072$

**Table 1.** Brainstem morphometric values in the African giant pouched rat irrespective of sex (n = 29)

Table 2. Sexual dimorphism in the body and brain morphometry in the African giant pouched rat (mean ± SEM)

Sex	Body weight [g]	Absolute brain weight [g]	Relative brain weight (%)	Nose-rump length [mm]	Absolute brain length [mm]	Relative brain length (%)	n Brain volume (mL)
Female ( $n = 15$ )	922.667 ± ± 65.571	$5.452 \pm \pm 0.088$	0.636 ± ± 0.049	336.087 ± ± 8.703	36.545 ± ± 0.242	11.038 ± ± 0.340	3.827 ± ± 0.141
Male (n $=$ 14)	1082.143 ± ± 46.174 <sup>*</sup>	5.928 ± ± 0.124**	$\begin{array}{c} 0.564 \ \pm \\ \pm \ 0.032^{\text{NS}} \end{array}$	$\begin{array}{l} 355.093 \pm \\ \pm \ 11.643^{\text{NS}} \end{array}$	$\begin{array}{l} 36.839 \pm \\ \pm \ 0.565^{\text{NS}} \end{array}$	$\begin{array}{l} 10.524\ \pm \\ \pm\ 0.376^{\text{NS}} \end{array}$	$\begin{array}{c} 3.614 \ \pm \\ \pm \ 0.094^{\text{NS}} \end{array}$

NS — non-significant difference (p > 0.05); \*significant difference (p < 0.05); \*\*highly-significant difference (p < 0.01)

were 999.655  $\pm$  42.622 g and 345.262  $\pm$  7.293 mm, respectively. Similarly, the mean brain weight and length of all the rats in absolute and relative terms were 5.682  $\pm$  0.086 g, 0.601  $\pm$  0.160%, 36.687  $\pm$  0.296 mm, and 10.764  $\pm$  1.322%, respectively. The mean brain volume obtained in the African giant pouched rat, irrespective of sex, was 3.724  $\pm$   $\pm$  0.087 mL. Table 1 shows the results of the weights and dimensions of the brainstem structures in all the 29 rats, irrespective of their sex.

With respect to sex, the relative brain weight was higher in the female than in the male rodent (Table 2) but the difference was not significant (p > 0.05). Similarly, there was no significant (p > 0.05) difference in the absolute and relative brain lengths or brain volumes between the two sexes, as shown in Table 2. The heights of the rostral colliculi in the male and female rats were 0.833 ± 0.223 mm and 0.627 ± 0.168 mm, respectively, while the heights of the caudal colliculi were  $0.820 \pm 0.220$  mm and  $0.942 \pm 0.252$  mm in the male and female rats, respectively. The sexual differences in the heights of these colliculi (Fig. 1) were not significant (p > 0.05). No significant sexual dimorphic value was obtained in the weights and dimensions of the mesencephalon and the pons, as shown in Tables 3 and 4. Furthermore, the difference in the weights, lengths, and volumes of the myelencephalon (Table 5) were not significant (p > 0.05).

The differences in body weight (Table 2), relative weight of mesencephalon (Table 3), and the length of pyramid (Table 5) were significant (p < 0.05) in male and female African giant pouched rats. The diameters of the rostral colliculi in the male and female rats were  $0.392 \pm 0.105$  mm and  $0.310 \pm 0.083$  mm, respectively, while the diameters of the caudal colliculi were  $0.334 \pm 0.089$  mm and  $0.468 \pm 0.125$  mm in the male and female rats, respectively.



Figure 1. Sexual dimorphism in morphometric values of the mesencephalic tectum in the African giant pouched rat.

**Table 3.** Morphometric values of the mesencephalon in the African giant pouched rat (mean  $\pm$  SEM)

Sex	Mesencephalon weight [g]	Mesencephalon weight as % of brain weight (relative weight)	Mesencephalon length [mm]	Mesencephalon length as % of brain length (relative length)	Mesencephalon width [mm]
Female ( $n = 15$ )	$0.573\pm0.030$	$10.425 \pm 0.522$	$5.182\pm0.328$	$14.273 \pm 1.005$	$6.577 \pm 0.594$
Male (n = 14)	$0.514\pm0.038^{\text{NS}}$	$8.675\pm0.585^{\text{NS}}$	$4.431\pm0.140^{\text{NS}}$	$12.061 \pm 0.407^{\text{NS}}$	$7.544\pm0.241^{\text{NS}}$

NS — non-significant difference (p > 0.05)

Sex	Pons weight [g]	Pons weight as % of brain weight (relative weight)	Pons length [mm]	Pons length as % of brain length (relative length)	Pons width [mm]	
Female ( $n = 15$ )	$0.162\pm0.009$	$0.575 \pm 0.154$	$3.462\pm0.201$	$2.132\pm0.570$	$1.679\pm0.043$	
Male ( $n = 14$ )	$0.179\pm0.012^{\text{NS}}$	$0.780\pm0.206^{\text{NS}}$	$3.464\pm0.161^{\text{NS}}$	$1.612\pm0.431^{\text{NS}}$	$1.707\pm0.037^{\text{NS}}$	

NS — non-significant difference (p > 0.05)

**Table 5.** Sexual dimorphism in the morphometric values of the myelencephalon in the African giant pouched rat (mean  $\pm$  SEM)

Sex	Myelencephalon weight [g]	Myelencephalon length [mm]	Myelencephalon volume [ml]	Pyramid length [mm]	Pyramid width [mm]	
Female (n $=$ 15)	$0.546 \pm 0.025$	$10.311 \pm 0.569$	$0.593\pm0.016$	$7.177 \pm 0.562$	$1.361 \pm 0.102$	
Male (n $=$ 14)	$0.599\pm0.028^{\text{NS}}$	$10.081 \pm 0.389^{\text{NS}}$	$0.579\pm0.019^{\text{NS}}$	$8.779 \pm 0.216^{*}$	$1.612 \pm 0.093^{**}$	

NS — non-significant difference (p > 0.05); \*significant difference (p < 0.05); \*highly-significant difference (p < 0.01)

tively. The sexual differences in the diameters of these colliculi (Fig. 1) were significant (p < 0.05). Also, the differences in absolute brain weights (Table 2) and pyramidal widths (Table 5) in the male and female rodents were highly significant (p < 0.01).

## DISCUSSION

This study has expounded the morphometry of the brainstem structures in male and female African giant pouched rats. As stated earlier in the methodology, all skulls were fixed in 10% buffered formalin for 3 days to enable brain extraction with minimal damage. Therefore, it is pertinent to mention that the morphometric result obtained was that of 3 days formalin-fixed brain samples. Mayhew et al. [11] recorded a volumetric shrinkage factor of 1.25 and an area shrinkage factor of 1.16 in the brain of dog fixed in 7% buffered formalin for 1–2 months. Cutts [6] recorded a mean shortening of 1.06% (shrinkage factor: 0.0106) in the length of isolated skeletal muscle of laboratory rats fixed in 10% formalin for 3 days. This implies that formalin has the property of causing minimal tissue shrinkage, the degree of which varies with duration of fixation. To the best of our knowledge, there is no available literature on the shrinkage factor of the brain of the African giant pouched rat, which we would have utilized in our results. However, the shrinkage expected in the present study, following 3 days fixation, may be insignificant.

The central nervous system is made up of the brain and spinal cord. The most rostral portion of the brain is the olfactory bulb, which lodges into the nasal compartment. On the other hand, the spinal cord terminates at the conus medullaries located at the level of the rump. Therefore, it was assumed in the study that while the nose represents the onset of the central nervous system in the African giant pouched rat, the rump represents its termination. Thus, the result of the brain length as a percentage of the nose-rump length (relative brain length) reflects, to a large extent, the length of the central nervous system occupied by the brain in the African giant pouched rat. The result of this relative brain length showed no significant difference in the male and female African giant pouched rats.

The sexual dimorphism in the body weight of the African giant pouched rat reported in this study corresponds with literature reports on the same species of rat by Henwood [8] and Nowak [13]. The finding is also similar to that of adult African white-tailed rats (*Mystromys albicaudatus*), as reported by Becker and Middleton [3], who stated that the males are significantly heavier than the females. Conversely, Byanet et al. [5] recorded a higher body weight in female grasscutters (*Thryonomys swinderianus*).

The sexual dimorphism in absolute brain weight reported in this study differs from that reported in the grasscutter by Byanet et al. [5] who observed that the female brain weighs more than the male brain in absolute terms. Similarly, Olopade and Onwuka [14], Onwuka et al. [17], and Olopade et al. [15] reported that female Red Sokoto goats, West African Dwarf goats, and West African Dwarf sheep, respectively, had heavier brain weights than their male counterparts in absolute terms; although in all of the above studies, the females had higher body weights. Conversely, Oto and Haziroglu [18] reported that the brain of the male donkey is significantly heavier than that of the female donkey in absolute terms. This is in agreement with the present result obtained on the brain of the African giant pouched rat. The study of Oto and Haziroglu [18] and the present study on the African giant pouched rat recorded higher body weights in male than female animals. This implies that absolute brain weight is directly proportional to body weight; hence the preference of relative brain weight to absolute brain weight in species comparison. The sexual dimorphism in brain weight may not mean that the sex with the heavier brain is more intelligent than the other. This is due to the controversy surrounding the correlation between brain weight and intelligence. Earlier reports [4, 20] suggested a relationship between brain weight and intelligence, but later findings by Gläscher et al. [7] emphasized that the internal structural complexity of the brain and interconnection of specific brain centres are the most important factors in the evolution of intelligence, and not the brain size.

Olude et al. [16] reported a neurocranial length of 3.33 cm and neurocranial volume of 5.06 mL in the African giant pouched rat. The neurocranial length is almost the same as the absolute brain length of 36.687 mm (or 3.669 cm) reported in the present study, while the neurocranial volume may be comparable with the absolute brain volume of 3.724 mL observed in the present study. Thus, neurocranial morphometry may be a good estimate of brain morphometry.

The brainstem morphometry in the African giant pouched rat observed in the present study did not show considerable sexual dimorphism. Aside from the diameter of the colliculi and the dimensions of the pyramids, all other brainstem structures measured with respect to sex showed no significant difference. The absence of any significant sexual difference in these brainstem structures in the African giant pouched rat implies that their respective functions may not be more efficient in either of the two sexes. The male rat showed a larger rostral colliculi while the female showed larger caudal colliculi. However, while the height differences in these colliculi were not significant, their relative diameters were significantly different, as observed in Figure 1. Baron et al. [2] reported that the size of the caudal colliculus reflects the hearing capacity of species better than any other brain structure. The rostral colliculus is an important centre in the visual pathway [12]. Therefore, in respect to the function of these colliculi in the visual and auditory pathway, it may be that the male African giant pouched rat has a relatively better visual sense than the female while the female rat may have a better acoustic sense than the male. The significantly higher value of the dimension of the pyramids in the male than female African giant pouched rat implies that the motor control of the hind limbs may be more efficient in the male than in the female rat.

## CONCLUSIONS

This study has established that the role played by sex in the morphometric values of the brainstem in the African giant pouched rat is minimal. The study has also provided, for the first time, baseline data on the absolute and relative weight, length, and volume of the intact brain of the African giant pouched rat with respect to sex, which has been lacking in the literature.

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