Comparative Anatomical Studies of the Cerebrum, Cerebellum, and Brainstem of Males Guinea pig *(Cavia porcellus)* and Rabbit *(Oryctolagus cuniculus).*

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Abstract

Comparative morphologic, morphometric and histological studies were carried out on the cerebrum. cerebellum and brainstem of ten adult male mammalian species namely the Guinea pig (Cavia porcellus) and Rabbit (Oryctolagus cuniculus). The animals were acclimatized for three days, weighed and lightly anaesthetized with chloroform, humanly sacrificed and perfused with physiological saline and Bouin's fluid. The brain were removed through midline incision through the skull and then fixed in Bouin's fluid, processed routinely and stained using Haematoxylin and Eosin stains. The brains were observed to be milky in colour and globular in shape in the two mammalian species though the size differed in the animals.

The mean body weight of Guinea pig and Rabbit were 454.00 ± 53.43g and 1393.00 ± 61.68g respectively. The results showed a statistical significant increase of the mean body weight, whole brain weight, weight of the cerebrum, weight of the cerebellum and weight of the brainstem in Rabbit compared to Guinea (P<0.05). The mean of length, width and thickness in cerebrum, cerebellum, and brainstem of Rabbit were showed to statistically higher when compared to Guinea pig (P<0.05). The result of the histological observation of the cerebral cortices (pyramidal cell layer, plexiform layer and granular layer which vary in size of layer and cellular population) and cerebellum shows similarities between the two species. The results from the present study can be used as a baseline

research data in comparative neuroanatomy for related rodent species.

Keywords: Comparative, Micro – morphology, Cerebrum, Cerebellum, Brainstem, Guinea pig, Rabbit

Introduction

Comparative anatomy is the study of similarities and differences in the ana-tomy of different species. Comparative anatomy has long served as evidence for evolution; it indicates that various organisms share a common ancestor. Also, it assists scientists in classifying organisms based on similar characteristics of their anatomical structures (Campbell et al., 2002. The guinea pig (Cavia porcellus), also called the cavy, is a species of rodent belonging to the family caviidae and the genus Cavia. Despite their common name, these animals are not in the pig family, nor are they from Guinea. They originated in the Andes, and earlier studies based on biochemistry and hybridization suggested they are domesticated descendants of a closely related species of cavy such as Cavia aperea, Cavia fulgida, or Cavia tschudii and therefore, do not exist naturally in the wild (Weir, 1974; Nowak, 1999). Their domestication began around 5000 B.C. and because of their popularity as pets and food, they are now globally distributed. They are indigenous to South America, with fossil records extending as far back as 9000 B.C. European colonization of South America led to their introduction as pets in Europe and ultimately, the world over (Morales, 1994: Vanderlip, 2003). Rabbit (Oryctolagus cuniculus), also called European, an old world, or a domestic rabbit is the only specie in its genus. The male is called buck and the female is doe, a young rabbit is a kitten or kit. The rabbit belong to the family leporidae, genus orycto-lagus and specie cuniculus. In Nigeria, it has the following local names, "zomo" in Hausa, "ehoro" in Yoruba, "ewii" in Igbo. Rabbits are ground dwellers that live in environment ranging from desert to tropical forest and wetland (Parker, 1990: Wilson and Reeder, 1993).

The cerebrum, the largest subdivision of the human brain, consists of a pair of cerebral hemispheres (Saladin, 2001). When viewed from the lateral aspect each cerebral hemisphere has three somewhat pointed ends or poles. These are the frontal pole rostrally the occipital pole caudally and the temporal pole that lies between the frontal and occipital poles and points forwards and somewhat down-wards, while the parietal lies laterally. A coronal section through the cerebral hemispheres shows that each hemisphere has three borders: Dorsomedial. ventrolateral. ventromedial. These borders divide the surface of the hemisphere into three large surfaces: Dorsolateral, Medial, ventral (Singh, 2009). The cerebellum (or small brain) lies in the caudally cranial fossa. In the human adult, the weight of the cerebellum is about 150g. The cerebellum has a superficial layer of grey matter, the cerebellar cortex. The cerebellum lies behind the pons and the medulla. It is separated from the cerebrum by a fold of dura matter called the tentorium cerebelli (Singh, 2009).

The brainstem consists (from above downwards) of the midbrain, the pons and the medulla. The midbrain is continuous, above, with the cerebral hemisphere. The medulla oblongata is continuous, below, with the spinal cord. Caudally, the pons and the medulla are separated from the cerebellum by the fourth ventricle (Singh, 2009). Midbrain, when viewed from the rostral aspect, two large bundles of fibers are seen, one on each side of the middle line. These are the Crura of the midbrain. The caudal aspect of the midbrain is marked by four rounded swellings. These are the Colliculi. One rostral and one caudal on each side (Singh, 2009). The pons shows a convex rostral surface, marked by prominent transversely running fibers. Laterally these fibers collect to form a bundle, the middle cerebellar peduncle. The anterior surface of the pons is marked, in the midline, by a shallow groove, the Sulcus basilaris, which lodges the basilar artery. On each side of the lower part of the pons there is a region called the Cerebellar-pontine angle. The medulla oblongata in the African grass cutter was conical in shape (Ajayi et al., 2011). It was located caudal to the pons. Its dorsal surface formed the lower boundary of the fourth ventricle lodged in a groove on the ventral surface of the cerebellum. Caudally, the medulla oblongata continued as the spinal cord with the foramen magnum as the boundary (Ajayi et al., 2011). The aim of the present work was to study the comparative micro - morphology of the cerebrum, cerebellum and brainstem of two mammalian species namely the Guinea pigs and Rabbits.

Materials and Methods Experimental Animals

A total of five adult male guinea pigs and five adult rabbits were used for this study. The animals were purchased from Sabo Gari market, Zaria. They were kept in a metal cage in the animal house, in the department of Human Anatomy, Ahmadu Bello University. Zaria. The animals were fed with vital feeds and water. The animals were acclimatized for three days before the commencement of the study.

Animal Sacrifice

The animals were weighed using weighing balance and anaesthetized lightly using chloroform. The animals were then sacrificed humanely and perfused through the common carotid artery with physiologic saline solution first, and then with Bouin's fluid. The whole brain was exposed from the rostral vault, and removed by gentle traction with scalpel blade, and was weighed using a digital scale. The components of the brain were then detached, weighed and recorded. The whole brain was fixed in Bouin's fluid.

Morphological Studies

The structural characteristics of the animals were examined; the weights of the animals were measured before sacrifice and the area of the head and the brain were examined with naked eyes and hand lens after sacrifice. This observation includes: shape size, borders, and weight of the brains. The presence of gyri and sulci were evaluated in each brain. The fixed brain tissues were processed routinely and stained using Haematoxylin and Eosin stain (Bancroft (H&E) and Gamble. 2008).

Statistical Analysis

Data obtained were expressed as mean \pm standard error of mean (SEM). Difference between group means was estimated using Students *t* – test using statistical package for social sciences (SPSS) version 22. A P value (≤ 0.05) was considered statistically significant.

Results

Morphologic Observations

The brain of the two species, the rabbits and the Guinea pigs were milky in colour, it shows major depressions separating the cerebrum from the cerebellum and other separating the cerebrum into two hemispheres, the shape of the cerebrum in the rabbits were triangular while diamond shape in the Guinea pig. Each cerebral hemisphere was large, and differentiated into four lobes: frontal, parietal, temporal and occipital. Both were devoid of prominent gyri and sulci. The olfactory bulbs were also prominent and visible from the dorsal view. The cerebellum was very distinct and secondary to the cerebrum in size. The cerebellar hemisphere was highly coiled with a distinct unpaired vermis; the paired flocculi, with its lateral accessory part, the paraflocculus, were also paired. The cerebellum was observed to be irregularly globular in shape. The cerebellum covers only the rostral part of the

medulla. The midbrain was composed of four corpora quadrigemina or colliculi. The pair of rostral ones is known as superior colliculi and the pair of caudal one's is called inferior colliculi (Fig 1). The Rabbits medulla was conical in shape but in guinea pigs, it was almost entirely covered by cerebellum (Fig 2).

On the ventral side of guinea pig brain, the paired optic nerves (CN II) were united at the optic chiasma and gave off optic tracts that ran on the rostral border of the tuber cinerium similar to rabbit. It was devoid of prominent gyri and sulci in both species.

Morphometric Study

The morphometric study showed the mean and standard error of mean of weight, length, width and thickness of the whole brain, cerebrum, cerebellum and brainstem in the Rabbits and Guinea pigs.

The results in table (1), showed the mean and standard error of mean of the total body weight, whole brain weight, weights of the cerebrum, weight of the cerebellum and weight of the brainstem. There was significant increase in the total body weight, whole brain weight, weight of the cerebrum, weight of the cerebrum, weight of the cerebrum, weight of the cerebrum, in Rabbits compared to Guinea pigs (P<0.05).

The results in table (2), showed the mean and standard error of mean of length in cerebrum, cerebellum, and brainstem. There was significant increase in the length of cerebrum, cerebellum, and brainstem in Rabbits compared to Guinea pigs (P<0.05).

The results in table (3), showed the mean and standard error of mean width in cerebrum, cerebellum, and brainstem. There was significant increase in the width of cerebrum, cerebellum, and brainstem in Rabbits compared to Guinea pigs (P<0.05).

The results in table (4) showed the mean and standard error of mean of thickness in cerebrum, cerebellum, and brainstem. There was significant increase in the thickness of cerebrum, cerebellum, and brainstem in Rabbits compared to Guinea pigs (P<0.05).

Microscopical Observations

The cerebral cortices in the examined two mammalian species were similar in terms of the structural composition as they are consisting of six distinguished layers. The first, plexiform or molecular layer consists of few cells. A dense population of small pyramidal cells and stellate cells make up the second layer called the external granular layer, the third layer, external pyramidal consist of medium pyramidal cells, the fourth layer or the internal granular layer consist of many stellate cells. The fifth layer or internal pyramidal cell layer consisted of large pyramidal cells, while the last layer called the multiform or fusiform layer contains numerous small pyramidal cells and cells of Martinotti, as well as stellate cells (Figs 3 - 6).

The cerebellar cortices in the two mammalian species are similar in term of their structural composition as they are consisting of three distinguished layers. The outer molecular (plexiform) layer, the inner granular cell layer. Between the two layers is the Purkinje cell or piriform layer. The Purkinje cells were seen to be conspicuously round in shape, with their nuclei located towards the periphery. The granular layer contained numerous small cells called granule cells. The molecular layer contained stellate and basket cells (Figs 7, 8).

Discussion

Morphologically, in this present study, the cerebral hemispheres of guinea pig and rabbit lacked prominent gyri and sulci. The absence of prominent cerebral hemisphere gyri and sulci placed the guinea pig and rabbit brains in the lissencephalic group (Greene *et al.*, 1976). A similar lissencephalic brain has been reported in African giant pouched rat by (Ibe *et al.*, 2014), in some

sciuromorphs and myomorphs by (Pilleri et al., 1984) and in hystricomorphs by (Dozo et al., 2004). Triangular shaped cerebral hemisphere was observed in the present study. Potter and Brueck (1958) reported a diamond- shaped cerebral hemisphere in guinea pig. The cerebral hemispheres of guinea pig and rabbit have four lobes: frontal, parietal, temporal and occipital. This is Similar to that of human (Singh. 2009). Olfactory bulbs observed in the present study were well developed from the dorsal view of the intact brain, although they are structures located on the ventral surface of the brain similar to that of African giant rat (lbe et al., 2014). In some mammals, such as elephants (Shoshani et al., 2006), the olfactory bulbs are small and therefore not visible from the dorsal view of intact brain; while in other mammals, such as whales (Marino et al., 2003), olfactory structures are absent.

Morphometrically, in the present study, the mean weights of body, brain, cerebrum, cerebellum, brainstem and whole brain of rabbits were larger than those of guinea pigs. The results of this study showed the brain weight of Guinea pigs and Rabbits to be less than figures reported for dog, cat, and porcupine, but larger than laboratory rat and sparrow (Eric, 2006) respective-

ly. Russel (1979) stated that there exists a relationship between brain size and intelligence. Earlier in history of neuroscience, Broca (1861) was of the opinion that brain size reflected intelligence. It was Broca himself that laid the grounds work for the modern view that the brain is a heterogenous collection of highly interconnected but functionally discrete system. Based on this, the rabbit and guinea pig can be said to be less intelligent than the dog, cat, porcupine (Eric, 2006), but more intelligent than laboratory rat and sparrow.

Conclusion

This study has shown the similarities and difference between the cerebrum, cerebellum and brainstem of two mammalian species; Guinea pigs and Rabbits. The result obtained from the present study can be used as research baseline for neuropharmacology and animal neuropsychiatry. It can also be used for comparative neuro-anatomy for other animals.

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	Guinea pig (n=5)	Rabbit (n=5)
Woight	Mean ± SEM	Mean ± SEM
weight	(g)	(g)
Body	454.00±53.43	1393.00±61.68*
Brain	3.30 ± 0.07	6.32 ± 0.17*
CER	2.43 ± 0.06	4.59± 0.22*
CEBM	0.43 ± 0.01	0.92± 0.03*
BRSM	0.31 ± 0.02	0.69 ± 0.01*
Whole Brain	2.99 ± 0.05	5.63 ± 0.18*

Table (1): Morphometric parameters of male Guinea pig and Rabbit

***P≤0.05 =** Statistically Significant; WT: weight; Min:

Minimum; Max: maximum; CER: cerebrum; CEBM: cerebellum; BRSM: brainstem;

Table (2): Length of Cerebrum, Cerebellum, and Brainstem of male Guinea pig and Rabbit

	Guinea pig (n=5)	Rabbit (n=5)	
Length (cm)	Mean ± SEM	Mean ± SEM	
Cerebrum	2.22 ± 0.35	2.72 ± 0.64*	
Cerebellum	0.89 ± 0.32	1.33± 0.48*	
Brainstem	1.30 ± 0.81	1.77 ± 0.18*	
* P≤0.05 = Statis	stically Significant		

Guinea nia (n=5)			Dabbit /n=/	5)	
Guinea pig and Rabbit					
Table (3): Thickness of Cerebrum,	Cerebellum,	and	Brainstem	male	of

Guinea pig (n=5)		Rabbit (n=5)	
Width (cm)	Mean ± SEM	Mean ± SEM	
Cerebrum	2.06 ± 0.24	2.46 ± 0.35*	
Cerebellum	1.44 ± 0.20	1.86 ± 0.35*	
Brainstem	0.87 ± 0.35	1.17 ± 0.09*	
* P≤0.05 = Statis	tically Significant		

Table (4): Thickness of Cerebrum, Cerebellum, and brainstem male ofGuinea pig and Rabbit

	Guinea pig (n=5)	Rabbit	(n=5)
Thickness (cm)	Mean ± SEM	Mean ± SEM	
Cerebrum	1.14 ± 0.14	1.70 ± 0.35*	
Cerebellum	0.76 ± 0.17	1.01 ± 0.58*	
Brainstem	0.53 ± 0.11	0.71 ± 0.20*	

*P≤0.05 = Statistically Significant

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Fig (1): Dorsal view of rabbit brain

Dorsal view of guinea pig brain



Fig (2): Ventral view of rabbit brain

Ventral view of guinea pig brain



Fig (3): Photomicrograph of the transverse section of cerebral cortex of Guinea pig showing: I Molecular layer, II: Outer Granular layer, III Outer pyramidal layer, IV Inner Granular layer, V Inner pyramidal layer. H&E stain. Mag. X100.



Fig (4): Photomicrograph of the transverse section of cerebral cortex of rabbit showing: I Molecular layer, II: Outer Granular layer, III Outer pyramidal layer, IV Inner Granular layer, V Inner pyramidal layer, VI Multiform layer. H&E stain. Mag. X100.

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Fig (5): Photomicrograph of the transverse section of cerebral cortex of Guinea pig showing: PC Pyramidal cells, PCN Nuclei of pyramidal cells. H&E stain, Mag. X250



Fig (6): Photomicrograph of the transverse section of cerebral cortex of rabbit showing: PC Pyramidal cells, PCN Nuclei of pyramidal cells. H&E stain, Mag. X250



Fig (7): Photomicrograph of the transverse section of cerebellar cortex of Guinea pig showing: PC Purkinje cells, ML Molecular layer, GC Granular cells. H&E stain. Mag. X 250.



Fig (8): Photomicrograph of the transverse section of cerebellar cortex of rabbit showing: PC Purkinje cells, ML Molecular layer, GC Granular cells. H&E stain. Mag. X 250.