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On the Structure of the Spleen in the African Giant Pouched Rat (*Cricetomys gambianus*, Waterhouse 1840)

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Abstract: African giant pouched rats are currently explored as laboratory model of haematological investigations but few detailed anatomical descriptions have been made of their spleen, limiting their use for such biomedical research. We studied the architecture of the spleen in six adult and clinically healthy African giant pouched rats by gross observation and light microscopy. The spleen was a long slipper-shaped and dorso-ventrally flattened organ lying on the left lateral surface of the abdominal cavity, medial to the lateral abdominal wall and lateral to the greater curvature of the stomach. Statistical analysis showed an average spleen weight of 3.31 g (± 0.54) and length of 6.44 mm (± 0.48). The spleen constituted 0.313% of the body weight and 2.069% of the nose-rump length. There was a very strong positive correlation between body weight and spleen weight (r = 0.958).

Key words: African giant pouched rat, red pulp, white pulp, megakaryocytes, subterranean habitat, histologically

INTRODUCTION

Rodents constitute the largest number of animals in the class Mammalia (Parker and Heswell, 1974). The high fecundity, relatively cheap cost of management, easy restrain, vast number of available antibodies, etc. have placed rodents on the zenith of laboratory model of biomedical research. The African giant pouched rat is a wild, subterranean rodent found in Africa including Nigeria (Rosevear, 1969; Ajayi, 1975). The potential of the African giant pouched rat as a laboratory model for biomedical research has not been fully exploited. This may be sequel to the dearth of published detail on the biology of the rat compared to, for example, the more widely used Wistar rat. However, efforts are ongoing to effectively domesticate them for several purposes including serving as a laboratory model (Olayemi and Adeshina, 2002).

The spleen is a vital haemopoeitic and immune organ. It is one of the most important immune organs of vertebrates and the principal peripheral lymphoid organ (Rooney et al., 2003; Balogh et al., 2004). It houses some of the body's lymphocytes and initiates cellular immune response and immunological defence through antibody production against blood-borne antigens (Nolte et al., 2002; Balogh et al., 2004). The lymphocytes are resident in the white pulp of the spleen. The spleen, through the red pulp plays a central role in the filtration of effete blood and foreign materials from the body. The red pulp also serves as a storage site for iron, erythrocytes and

platelets and has been implicated to be haemopoeitic in neonatal rodents (Cesta, 2006). The macroscopic features and size of the spleen varies with species (Cesta, 2006).

As a preliminary to the evaluation of pathological conditions, it is necessary to establish the normal structure of the spleen in any given specie otherwise, morphological changes associated with specie and body size may be confused for toxic-induced lesions.

For the Muridae family, several gross and light microscopic studies on the normal structure of the spleen have been undertaken on the brown rat (Furrianca et al., 2008), ground squirrel (Shivacheva and Khadzhiolov, 1987) and mole rat (Kotze et al., 2006). Although, the amount of study on the anatomy of the African giant rat in Nigeria is gradually increasing (Ogwuegbu et al., 1983; Oke et al., 1988a, b; Oke and Aire, 1989, 1990; Nzalak et al., 2005; Akinloye et al., 2007; Ali et al., 2008; Olude et al., 2009; Ibe et al., 2010), the regional anatomy of the spleen suffers relatively from lack of research attention. Consequently, the aim of the present paper was to provide an overview both by gross and light microscopy of the anatomy of the spleen in the African giant pouched rat. The objectives of the study were as follows: firstly to describe the gross and histo-morphology of the spleen of the African giant pouched rat this has hitherto not been reported. Secondly, to establish the morphometric features of the spleen. Thirdly, to compare such data with those from other rodents that has been reported. This is

the first of a series of reports in which the splenic structure of this rat will be characterised in detail, both in qualitative and quantitative terms.

MATERIALS AND METHODS

About 6 spleens with no visual sign of infection obtained from clinically healthy adult African giant pouched rats were used for the present study. The animals were captured from the wild in Kaduna State, Nigeria using locally made traps. They were transported by road to the animal house in the Department of Veterinary Anatomy, Ahmadu Bello University Zaria. The animals were acclimatized for 3 weeks before commencement of the experiment during which they were physically examined under careful restraint. Water and feed were provided ad libitum and they were maintained under constant environmental conditions. Following sedation with 20 mg kg⁻¹ Thiopental sodium, i/p, the animals were weighed and euthanized with a lethal dose of same drug. The spleen was exposed after an exploratory laparotomy. Its orientation and relationship with other abdominal viscera were established. Since the method of euthanasia employed caused death by blood accumulation in the spleen due to the hypotensor effect of the anaesthetic (Hardman et al., 1996), it employed the method of Galindez et al. (2006) to drain excess blood from the exteriorised spleen before the dimensions and weights were obtained using a vernier calliper (MG6001DC, General Tools and Instruments Co., New York; sensitivity: 0.01 mm) and electronic sensitive balance (Mettler balance P 1261, Mettler instrument AG. Switzerland; sensitivity: 0.001g), respectively. Bouin's fixative was perfused through the hilus to preserve the tissues for histological study. The spleens were thereafter immersed in the same fixative.

After dehydration in ascending concentrations of alcohol, the tissues were cleared in xylene, infiltrated with molten paraffin wax (BDH Chemicals Ltd. Poole, England) at 60°C, blocked in paraffin according to standard procedures (Kiernan, 1990) and labelled.

Coronal and saggital sections of 5 µm each were cut with a Jung rotary microtome (Model 42339, Berlin, Germany) and mounted on glass slides using DPX mountant. The sections were stained with haematoxylin and eosin (H/E). The tissue slides were observed under a light microscope (Olympus-XSZ107BN, Hamburg, Germany) connected to a digital camera (Olympus DP70; Olympus).

RESULTS AND DISCUSSION

Gross morphology: The spleen was a long, slipper-shaped, dark red coloured organ, lying on the left lateral surface of the abdominal cavity, medial to the lateral



Fig. 1: Dorsal view of the spleen in the African giant pouched rat

Table 1: Summary of result on body and spleen morphometry in the African giant pouched rat

n = 6	Min.	Max.	Mean±SEM
Body weight (g)	950.00	1250.00	1065.00±52.20
Spleen weight (g)	1.95	5.11	3.31±0.540
N-R length (mm)	295.00	340.00	313.00±8.000
Spleen length (mm)	5.40	8.00	6.44±0.480
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N-R: Nose-Rump

abdominal wall and lateral to the greater curvature of the stomach. The spleen was dorso-ventrally flattened and was bigger in one pole (Fig. 1). The body weight, spleen weight, nose-rump length and spleen length in 6 adult African giant pouched rats were shown in Table 1. Statistical analysis showed a very strong positive correlation between body weight and spleen weight (r=0.958). There was also a positive correlation between nose-rump length and spleen length (r=0.460). The spleen constituted 0.313% of the body weight and 2.069% of the nose-rump length.

Histology: The coronal sections of the spleen had an elongated oval shape. The spleen was delineated into the splenic capsule (Fig. 1a) and the parenchyma (Fig. 1c). Projections of the capsule into the parenchyma in the form of splenic trabeculae were evident (Fig. 1b). Parenchyma of the spleen was composed of the red pulp (Fig. 2d) separated from the white pulp by a marginal zone (Fig. 2c). Within the white pulp, the lymphoid follicles composed of a pale staining germinal centre (Fig. 2a) and lymphocytes (Fig. 2b) where separated from the marginal zone by a septum (indicated with an arrow in Fig. 2). Blood vessels were numerous in the germinal centre. Very few megakaryocytes were observed in the red pulp of one spleen.

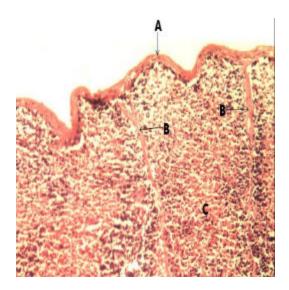


Fig. 2: Transverse section of the spleen of the African giant pouched rat, illustrating A: splenic capsule, B: splenic trabeculae, C: splenic parenchyma (HE x100)

Grossly, the spleen of the African giant pouched rat differed from that of other rodents. The spleen is larger at one pole of its longitudinal axis compared to the other. It is also slipper-shaped. This is unlike the spleen of the mice and rats which is uniform along the longitudinal axis (Cesta, 2006) and has the shape of an elongated triangle. The spleen is bean shaped in the alligator (Rooney et al., 2003) and guinea fowl (Onyeanusi, 2006). It has a shape of an elongated triangle in the musk shrew (Fukuta et al., 1982) and a more regularly triangular shape with rounded, blunt apices in the opossum (Dideiphis virginiana) (Cutts and Krause, 1982). The ratio of splenic weight to body weight in the African giant pouched rat is 0.313%. According to Losco (1992), the value in Wistar rats is 0.2%. The higher spleen size in the African giant pouched rat is a compensation for its subterranean habitat characterized of low oxygen tension as the spleen conserves more blood than that of the Wistar rat (Fig. 3).

Splenic capsule is made of connective tissue and smooth muscles, the relative amount of which varies with species (Brown and Dellmann, 1976). In the present study, distinct smooth muscle fibres were not observed in the splenic capsule-trabecular system. This is in accordance with the report of Valli *et al.* (2002) who stated that splenic capsule and trabeculae in rodents are not highly musculated and thus, do not undergo rapid contraction. Conversely, muscular tissues are predominant in the capsule-trabecular system of the hairy armadillo (Galindez *et al.*, 2006). The septum observed in the present study permits migration of lymphocytes from the white pulp into the marginal zone (Fukuta *et al.*, 1982).

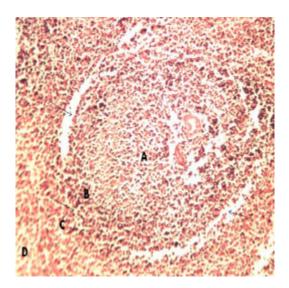


Fig. 3: Transverse section of the splenic parenchyma in the African giant pouched rat, illustration A: germinal centre, B: lymphocytes, C: marginal zone, D: red pulp. Arrows indicate the septum (HE x100)

According to Cormack (1987), the spleen in adult humans can be reactivated to carry out haematopoiesis under conditions such as severe haemolytic anaemia. However, extra medullary haematopoiesis is common in rodent red pulp especially in foetal and neonatal animals (Cesta, 2006). Chen and Weiss (1980) observed megakaryocytes in the splenic red pulp of the mouse while Zidan et al. (2000) observed megakaryocytes in the splenic red pulp of camels with the number of the megakaryocytes decreasing with age. The observation of few megakaryocytes in the present study is an indication of an on-going extramedullary haematopoiesis. However, since the splenic megakaryocytes are active in thrombocytopoiseis, resulting in platelet production, irrespective of age (Zidan et al., 2000), the observation of megakaryocytes in the spleen of the African giant pouched rat in the present study is indicative of thrombocytopoiseis. Thus, platelet formation in the spleen of adult African giant pouched rat is not sequel to a pathological disorder but is a normal function that is reduced in adult rats.

CONCLUSION

Histologically, the splenic capsule was distinct from the parenchyma which was well delineated into a red pulp and white pulp demarcated by a marginal zone. Few megakaryocytes, indicative of platelet production were observed in the splenic red pulp. The high spleen size in the African giant pouched rat, indicative of effective blood conservation is a compensation for their subterranean habitat characterized of low oxygen tension.

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