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Chromosomal Numbers in African Giant Rat (*Cricetomysgambianus*, Waterhouse-1840)

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Abstract: ***Background:*** *Karyotypic studies were carried out on the African giant rat, Cricetomysgambianus, Waterhouse-1840 with the aim of determining its chromosome diploid numbers and autosomal fundamental numbers.*
Methods*: The chromosomes were prepared from the conventional bone marrow of two (2) African giant rats – a male and a female treated intra-peritoneally with 2 ml of 0.04% colchicines for 3 hours. Chromosomes in well spread mitotic metaphase cells were counted and measured using KaryoType computer software. Chromosomal numbers were identified. Ideograms were also constructed from the measurements. Data were collected and analysed using SPSS version 20.*
Results*: A diploid chromosome number of 2n = 80 with an autosomal fundamental number (NFa) of 66 and 95 were obtained for the species of C. gambianus used in this study. The X chromosomes were medium-sized metacentric and small acrocentric while the Y chromosome was small acrocentric.*
Conclusion*: Cricetomysgambianus was found to have an identifiable autosomal diploid number,The findings resembled those in Benin, Senegal, Niger, Cameroun and other countries.*
Keywords*: African giant rat, Cricetomysgambianus, Chromosome, Number, Autosomal fundamental number.*

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I. Introduction

The African Giant rat (AGR) also known as Gambian pouched rat belongs to the order *Rodentia*, Suborder *Myomorpha*, family *Cricetidae*, subfamily *Cricetomyiane* and genus *Cricetomys.*[1] It is a wild rodent consumed by the rural population in Nigeria. Two species have been recorded in Nigeria, *Cricetomysemni* and *Cricetomysgambianus*, Waterhouse-1840.[2]Other Gambian species exist in South Africa and they include *Cricetomysgambianusadventor, C. gambianusselindensis*and *C. gambianuscunator. Cricetomysemni* is distributed naturally in the rain forest zone and is not associated with human habitation. It is less common than the Gambian Giant rat.[2]

The African giant pouched rat, because of its exceptional size and other interesting attributes, is an economically important rodent within Africa. It is one of the most common mammals exploited as bush meat [3,4,5] and has been trained to aid in the detection of landmines [6] and also in the medical diagnosis of pulmonary tuberculosis.[7]

Several publications, employing alternative techniques such as karyotyping of the African Giant rat;[8,9,10,11] plasma biochemical properties of the African Giant rat;[12,13] Stereological estimation of the cerebral layers of African Giant rats;[14] multivariate craniometry[15] anatomical and histological studies of the digestive system of the African Giant rat;[16]morphologic, morphometric and histologic studies of cerebellum and forebrain of the African Giant rat;[17] morphometric studies of the cerebellum and forebrain of the African Giant rat;[18]morphometric characterization of the African Giant rat (*Cricetomys*Waterhouse 1840) in the forest zone of south western Nigeria;[19] weight assessment of some accessory digestive organs in the adult African pouched rat;[20] gross anatomical, histological and histochemical studies of the oesophagus of the African Giant rat;[21] histological and histochemical studies of the colon of the African rat [22] and gross anatomical aspect of gastro-intestinal tract of the wild African giant rat - *Cricetomysgambianus,*[23] have attempted to provide additional information useful for characterization of the various giant pouched rat species. However, the taxonomic impact of these studies has been of restricted importance because they were conducted on limited specimen collections, underscoring the need for more investigations covering the entire range of these rodents.

The use of molecular methods to provide more insight into the taxonomy and phylogeny of *Cricetomys*was recommended. Preliminary molecular studies involving this genus have helped to clarify its position and relationships with regard to other groups within the rodent superfamily *Muroidea*. [24]

In Benin, a list of rodent species has been published, but uncertainties remain about the taxonomic status of specimens from several genera, such as *Cricetomys, Tatera, Mastomys* or *Mus,* due to the absence of cytotaxonomic data. [25] Some chromosomal data also exist for the rodents of Benin, concerning the genera *Cricetomys*,[9]*Tatera*,[9,26]*Arvicanthis.*[27, 28]

Until recently karyological investigations on rodents of Senegal have mainly focused on *Gerbillidae*, and particularly the genera *Tatera*and *Taterillus.* These karyological studies made it possible to distinguish two sibling species of *Taterillus,* namely *T. gracilis*, with a diploid number of 2n = 36/37, and *T. pygargus*with (2n = 22/23), [29,30,31] and to characterize the two species of *Tatera, T. gambiana*(2n = 52)and *T. guineue*(2n = 50).[29,32, 33] In the family *Muridae*, data on the two previously known species of *Mastomys, M. erythroleucus*(2n = 38) and *M. huberti*(2n = 32) have been published,[34] but recent extensive studies of *Mastomys* genus in Senegal revealed the presence of a third species, *M. cfnatalensis,* morphologically indistinguishable from *M. huberti,* and having the same diploid number, but with distinct ecological preferences and a specific autosomal fundamental number (NFa = 54 versus 44) for *M. huberti.*[35,36,37]

II. Materials And Methods

This study was conducted on 2 adult African Giant rats (*Cricetomysgambianus,* Waterhouse-1840) of both sexes that were captured alive in the wild around Zaria City, Kaduna State, Nigeria using a local metal cage traps without inflicting injuries on them. They were housed in customized laboratory rat cages in the animal house of the Department of Human Anatomy, Faculty of Basic Medical Sciences, College of Health Sciences, Ahmadu Bello University, Zaria, Nigeria and fed with fruits, groundnut pellets and water was given *ad libitum* for a week prior to commencement of study.[38,39,40]

The rats were euthanized using anaesthetic chloroform in a confined container and weighed using a balance (EMPEROR model p.1210), made in Chandler, Arizona, United State of America (USA), with a sensitivity of 0.1 g. The live specimens were taken to the laboratory and injected intraperitoneally with 2 ml of 0.04% colchicine solution) prepared with sterile physiological saline. After a period of 3 hours of injection, the specimens were sacrificed using cervical dislocation.

Ethical approval and permission were obtained from Faculty of Basic Medical Sciences, College of Health Sciences, Ahmadu Bello University, Zaria, Ethics Committee for conducting the study.

Dividing cells were obtained from the marrow of the femur and tibia, dissected out in accordance with the methodology described by.[41,42,43] Both ends of the femur and tibia were cut open, and a hypodermic needle attached to a syringe containing 1 – 1.5 ml of freshly prepared and pre-warmed (37^{o}C) hypotonic buffer (0.55% KCl) was inserted. The marrow was flushed out into a 15 ml centrifuge tube.

Freshly prepared fixative (3:1 methanol – glacial acetic acid) was added drop wise, with quick agitation after each drop to re-suspend the cells. A total of 2.0 – 2.5 ml of the fixative were added.

From a height of about 1 metre, two or more drops of the cell suspension were allowed to fall on the slides uniformly. The slides were blown quickly across its length and placed on a slide warmer set at 60^{0}C, and were allowed to dry for about 24 hours before staining. The slides were flame dried for normal metaphase cells and air dried completely before staining.[44]

Giemsa powder (0.5 g) was dissolved in 33 ml glycerol and put in Erlmyer bottle in a dark compartment overnight. It was heated in a water bath set at 60^{o}C for 2 hours and was allowed to cool, after which 33 ml of methanol was added. This solution was stored in an amber colouredbottle as the stock Giemsa stain. G-bands were performed according to Seabright. [45] The slides were stained for 20 minutes, and rinsed in distilled water to remove the stain.

The chromosomes of an African giant rat were arranged in order of decreasing size, and the karyotype of*Cricetomysgambianus*were arranged in accordance with the report of Granjon*et al*. [8]

All statistical analyses were performed on an SPSS version 20.0 software package (SPSS Inc., Chicago, Illinois, USA). Data were presented as Mean ± SEM. One way analysis of variance (ANOVA) was used to compare the mean differences. P-values of less than 0.05 were considered to be statistically significant.

III. Results

The karyological study of the two individuals of *C. gambianus*revealed an autosomal diploid number within the species as 2n = 80 as shown in Table 1.

Table 1: Specie, sex, diploid number (2n),autosomal fundamental number (NFa), and morphology of the sex chromosomes of the *Cricetomysgambianus*.

Specie	Sex	2n	NFa	Sex chromosome
Cricetomysgambianus	M	80	88	X= MSSM and Y= Small A
	F	80	91	X=LSM and X=MSM

X=X chromosome,Y= Y chromosome, MSSM= Medium-sized submetacentric, A= Acrocentric, LSM= Large-sized metacentricandMSM= Medium-sized metacentric

Plate I shows a mitotic metaphase chromosomal spread of male *C. gambianus* specie. Figure 1 shows the karyogram was composed of 2n = 80, NFa = 88 with 2 pairs of metacentric, a pair of submetacentric, 2 pairs of acrocentric and 35 pairs of telocentric chromosomes. The X chromosome was medium-sized and submetacentric, and the Y chromosome was the smallest and acrocentric.

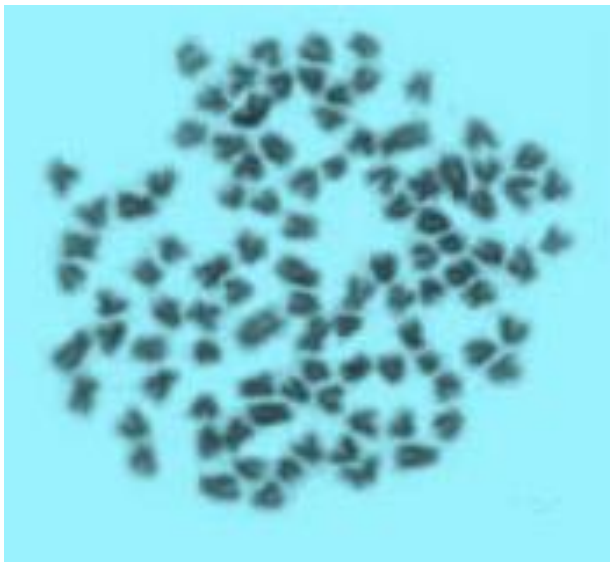

Plate 1 : Mitotic chromosomal spread of a male *C. gambianus* X 100 magnification using Giemsa stain

Figure 1: Karyotype of a male *C.gambianus*, 2N = 80, NFa = 88

Plate II shows a mitotic metaphase chromosomal spread of another female *Cricetomysgambianus* and Figure 2 shows the karyotype with a diploid number 2n = 80, NFa = 91, 5 pairs of metacentric, 1 submetacentric, 1 acrocentric and 33 pairs of telocentric chromosomes. The X chromosomes appeared as large and submetacentric and medium-sized and metacentric chromosomes.

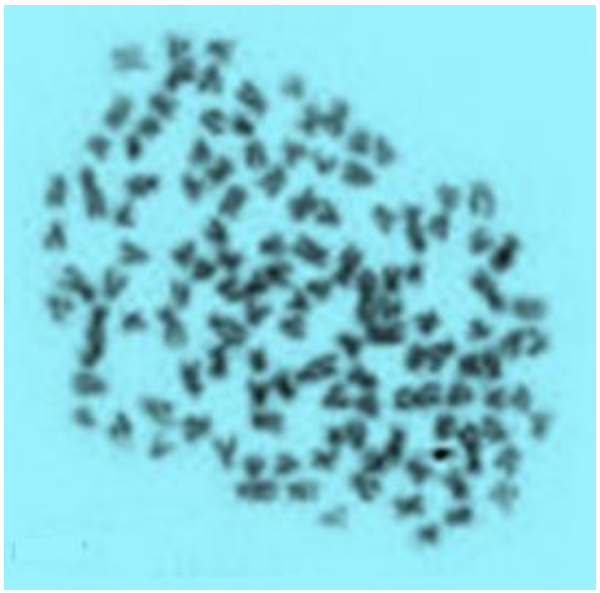

Plate 2: Mitotic chromosomal spread of a female *C. gambianus* using Giemsa stainX 100 magnification

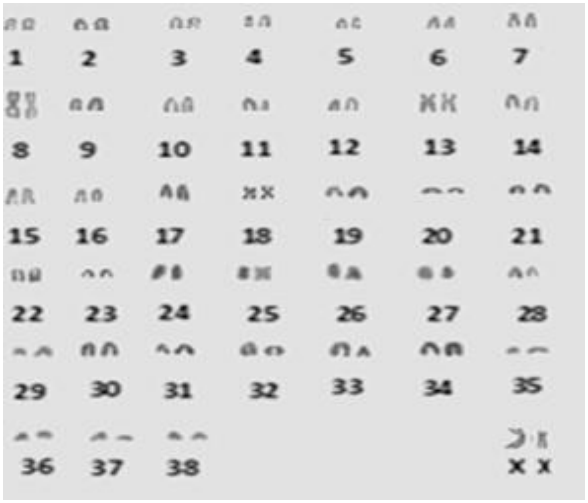

Figure 2: Karyotype of a female *C. gambianus*, 2N = 80, NFa = 91

IV. Discussion

In the present study, the autosomal fundamental number (NFa) of *C. gambianus* was found to be from 88 to 91 which is similar to those of *Arvicanthisrufinus*74[46] and *Lemniscomysbellieri*60-78; but disagree with those of *Taterakempi* 64 [26] and *Taterillusgracilis*44, [10]*Mastomysnatalensis*54, [28]*Myomysderooi*34 [47]and*Rattusrattus*58. [48]

The result of the study suggests that species with similar G-banding patterns are also similar morphologically, but major changes in the karyotype, as shown by G-banding, did not cause morphological change. [49] The closeness of the chromosomal number of the species could be due to ancestral origin because different types of chromosomal change may be found in groups of species which could plausibly have a common ancestry. [50]

The results are also quite similar to what Granjon*et al.*[8] found in Senegal, with the exception of one of the two metacentric autosomal pairs in the Lanta karyotype, which appear larger than in the karyotype from Senegal. In addition, the results differ from what was previously found in Benin by Codjia*et al.* [9]in which, 2n = 82, NFa = 88, from wild (N = 2, from the south of the country) and captive-bred (N = 3) specimens. The findings also differed with the published data provided by Matthey *et al.* [51] who found 2n = 78 for a *C.gambianus* of unknown origin.

The findings also differed with that published data provided by Matthey *et al.* [51]who found 2n = 78 for a *C.gambianus* of unknown origin. This is also differed with other species from same *muridae* family such as *Taterakempi* 48,[26]*Taterillusgracilis*39, [10]*Lemniscomysbellieri*56, *Mastomysnatalensis*32,[29]and*Rattusrattus*38. [48]

V. Conclusion

The present study also showed that the cytotaxonomic identification of the chromosomal patterns of *C. gambianus* resulted in a clear way of differentiating this genus from other genus within its family and from variable rodents within the genera.

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