Cytogenetic Study of Three Species of Rodents from Ile-Ife South-Western Nigeria

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Abstract
Cytogenetic analysis is one major tool useful for unravelling rodent diversity. Although in Nigeria interesting rodent diversity is currently being uncovered through molecular analyses, the cytogenetic aspects of this diversity are still largely unknown. In this study diploid number and chromosome morphology viewed on Giemsa-stained slides for three rodent species trapped in Obafemi Awolowo University Campus are reported. One of the specimens was also sequenced for Cytochrome b to assist in making accurate taxonomic designations. To our knowledge, chromosomal information is presented for two of these small mammal species (*Cricetomys gambianus*, *Cavia* spp.) for the first time from populations within South Western Nigeria. The diploid numbers detected in this study for *Cricetomys gambianus* (2n=64) differ from diploid numbers published for these species from other locations in previous studies and this raises a possibility of yet to be identified rodent diversity in the present area of study. Cytogenetic data combined with other lines of scientific evidence will help us appreciate and understand more the bountiful diversity of small mammals across Nigeria.

***Keywords**:* Biodiversity, Chromosome, Chromosome morphology, Cytogenetics, Rodent, Systematics.

Introduction
Information on African rodents is scattered over wide areas with the occurrence of sibling and cryptic species. As a result, the taxonomic revision of many genera is far from completion (Corti *et al*., 2005). About 386 species of rodents have been reported in Africa (Musser and Carleton, 1993). In the later part of 20th century, new disciplines such as genetics, cytogenetics, molecular genetics, and morphometrics have been developed and employed in the taxonomic studies of Africa rodents (Corti *et al*., 2005).

Rodents are one of the most successful mammal groups and are distributed almost throughout the world (Vaughan *et al*., 2000). African rodents are useful in biodiversity studies, playing key roles for instance in the food chain and in seed dispersal (Nyiramana *et al.,* 2011, Aliyu *et al.,* 2014). Rodents are also of agricultural and epidemiological importance. They have been implicated as crop pests (*Mastomys natalensis*; Mulungu *et al.* 2010) and also in transmitting deadly diseases such as plague (Kilonzo *et al.,* 2005) and Lassa fever (Lecompte *et al.,* 2006).

The intraspecific chromosome variation of African rodents which sometimes occurs within populations indicated the requirement of careful cytogenetic studies. It is generally accepted that the great diversity in rodents is related to their fast rate of chromosomal rearrangements. As a result, karyotype descriptions constitute the primary tool for rodent species identification (Fadda *et al.*, 2001; Corti *et al.*, 2005). Chromosomal data have the potential to reveal both structural and functional homologies among taxa, which can be utilized in phylogenetic and taxonomic investigations (Dobigny *et al*., 2004).

Karyotypic information has been reported for rodents from Obafemi Awolowo University Campus (Ashiru 1987; Akintoye 2000; Akintoye & Awopetu 2005a & b). But this study seeks to provide cytogenetic data for three species of these rodents that are available on Obafemi Awolowo University Campus.

Methods

The rodents were trapped from Obafemi Awolowo University campus (N07° 30' E04° 32') using locally fabricated live-traps made of steel. A preliminary identification of the animals, based on external morphology, was made by using the key provided in Rodents of West Africa by Rosevear (1969). Direct treatment of bone marrow cells was used for karyotype analysis, following the standard method in accordance with Patton (1967). Each live animal was intraperitoneally injected with 0.02 % colchicine at 0.01 ml/gram of body weight to arrest the cell division at the metaphase stage. After about two hours, the animals were sacrificed by etherization and bone marrow cells flushed out of the humerus and femur bones. The cell suspension was incubated in hypotonic solution (Potassium chloride) for 20 minutes at room temperature. The bone marrow cells were then fixed with freshly prepared fixative (3:1 methanol/glacial acetic acid). A total of 10 to 20 slides were prepared for each specimen karyotyped, and at least 30 well-spread metaphase cells from each preparation were analyzed. We used the chromosome nomenclature of Zima and Kral (1984). Chromosomes were designated as metacentric, submetacentric, subtelocentric and acrocentric according to the position of the centromere. The chromosome number obtained for each specimen was compared with reports from other studies in literature for identification and verification of homology.

Results

Table 1 shows the number of metaphase cells, range of diploid numbers, modal diploid number and fundamental number of autosomes of studied species. Representative spreads for all species are shown in Figure 1-3b. The karyological results of each species studied are presented as follows:

Karyotype of female Cricetomys gambianus

A diploid number (2n) of 64 and fundamental number (FN) of 64 were obtained for *Cricetomys gambianus* (Waterhouse, 1840). The karyotype for the female *Cricetomys gambianus* is shown in Figure 1. The karyotype of the female *Cricetomys gambianus* consists of 32 pairs of uniarmed autosomes. The karyotype includes 32 acrocentric autosome pairs. The sex chromosomes were not well differentiated.

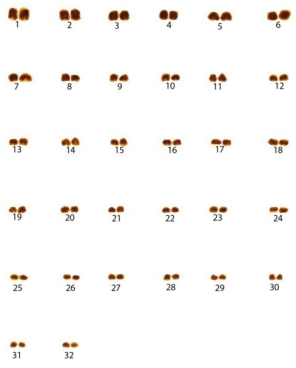
Table 1: Diploid and fundamental numbers for three rodent species captured in the study.

Species	Locality sampled	Number of spreads	Diploid number	Fundamental number (FNa)	Diploid number from other studies	Accession Number
Cricetomys gambianus	OAU*	200	64	64	80 (Corti *et al.,* 2005; Dobigny *et al.,* 2002; Granjon *et al.,* 1992)	KT232252
Rattus norvegicus	OAU	200	42	Male=52, Female= 62	42 (Yigit *et al.,* 1998)	Not sequenced
Cavia spp	OAU	205	48	54		Not sequenced

*'OAU' = Obafemi Awolowo University

Karyotype of male Cavia spp.

The diploid number obtained for *Cavia* spp. is 2n=48. The karyotype of the male *Cavia* spp. (Linnaeus, 1758) consists of 2 pairs of biarmed autosomes and 21 pairs of uniarmed autosomes. Autosomes consist of 4 metacentric/submetacentric and 42 acrocentric chromosomes (Figure 2). The X chromosome is metacentric while the Y chromosome is acrocentric. The fundamental number (FN) is 54.



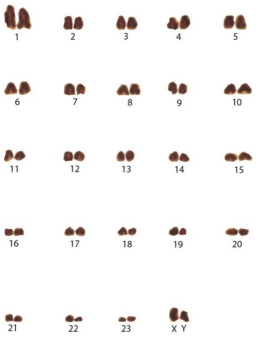
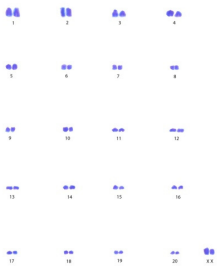


Figure 1: Karyotype of Female *Cricetomys gambianus.* Figure 2: Karyotype of Male *Cavia spp*



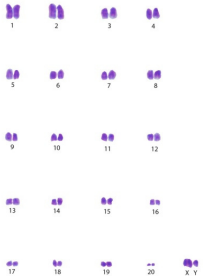


Figure 3a: Karyotype of Female *Rattus norvegicus*. Figure 3b: Karyotype of Male *Rattus norvegicus.*

Karyotype of female Rattus norvegicus (Berkenhout, 1769)
The diploid number of chromosomes is 2n=42. The karyotype of the male *Rattus norvegicus* (Berkenhout, 1769) consists of 10 pairs of biarmed autosomes and 10 pairs of uniarmed autosomes (Figure 3a). Autosomes consist of 20 metacentric/submetacentric and 20 acrocentric chromosomes. The X chromosome is acrocentric. The fundamental number (FN) is 62.

Karyotype of male Rattus norvegicus
The diploid number of chromosomes is 2n=42. The karyotype of the male *Rattus norvegicus* (Berkenhout, 1769) consists of 5 pairs of biarmed autosomes and 15 pairs of uniarmed autosomes (Figure 3b). Autosomes consist of 10 metacentric/submetacentric and 30 acrocentric chromosomes. The X and the Y chromosomes are acrocentric. The fundamental number (FN) is 52.

Discussion
A karyotype of 2n = 64 was discovered for *Cricetomys gambianus* in this study. This specimen was also sequenced for Cytochrome b to assist in making accurate taxonomic designations. This number is quite different from those published from this species from Senegal (2n = 80, FNa = 80; Granjon *et al.,* 1992), Benin (2n = 82, FNa = 88; Codjia *et al.*, 1994) and Niger (2n = 79-80, FNa = 82; Dobigny *et al.,* 2002). Thus chromosome numbers recorded from other parts of Western Africa for this species (2n = 79-82) are quite different to that now documented from southwestern Nigeria. The genus *Cricetomys* has been found to be quite variable across Africa, with Olayemi *et al*. (2012) discovering that there are at least 6 species within the genus where it was thought that there were only about 2 to 4 (Genest-villard, 1967; Musser & Carleton, 2005). This disparity in chromosome number could be a reflection of the diversity within the genus even below the species level. Although because of the quality of the metaphase spread obtained for this specimen, the possibility of the wide disparity in the diploid number obtained in this study resulting from some inconsistencies in metaphase chromosome preparation cannot be totally ruled out.

The diploid number obtained for the *Cavia spp.* in this study is 2n=48. To our knowledge, chromosomal information is presented for this rodent species for the first time from populations within South Western Nigeria. Despite extensive search of literature, there was no report to compare the result obtained in this study with and the karyotype information we have is not sufficient for accurate taxonomic designation of the specimen to species level. Ohno *et al.* (1961); Fernandez and spotorno (1968) and Dunnum and Salazar-Bravo (2006) reported a diploid numbers (2n = 64) in *Cavia tschudii* and *Cavia porcellus.*

The *Rattus norvegicus* specimens in our study have a chromosome number of 2n = 42. Aro (2014) also found this same number of chromosomes for the same species on the OAU campus in Ile-Ife, South West of Nigeria. Yigit *et al*. (1998) reported *Rattus norvegicus* with the diploid number 2n = 42 from Turkey. Our karyological findings are consistent with findings reported by the Committee for a standardized karyotype of *Rattus norvegicus* (1973), Yong (1969), Diaz de la Guardia (1981), Yosida (1973), Gamperl (1980) and Cao and Tran (1985).

In summary, our study present a karyotype of 3 rodent species, 1 of them –*Cricetomys gambianus* – had diploid numbers different from what has been recorded in past studies. This chromosomal variation in *Cricetomys gambianus* provides indication that there is quite an amount of diversity in Nigeria that remains to be described. A case has been established for higher rodent diversity than hitherto known from south western Nigeria (Nicolas *et al.,* 2010a & b; Olayemi *et al,.* 2012). Further research combining cytogenetic and molecular data with morphological details will help us understand more deeply the rodent diversity involving populations from the present study area and Nigeria in general.

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