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# Chromosomal Studies of the African Pygmy Hedgehog, *Atelerix albiventris* (Wagner, 1841)

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**Abstract**

*Chromosomal studies were carried out on samples of male and female African Pygmy hedgehog, Atelerix albiventris for the purpose of cytogenetic characterization and validation with the only existing report. Metaphase chromosomes were prepared from the bone marrow treated with 0.05% colchicine; well spread chromosomes were counted directly from resulting slides and photographed. A diploid chromosome number (2n = 48) was observed, with the Fundamental Number; FN being 93 in the male and 94 in the female. Autosomal fundamental number, FNa, was 90 in both sexes. The karyotype consisted of 14 pairs of metacentric, 6 pairs of acrocentric, 2 pairs of submetacentric and a pair of telocentric chromosomes. The X chromosomes were acrocentric while the Y chromosome is a micro-chromosome. From the karyotypic analysis in this study it was concluded that the diploid chromosome number of A. albiventris is consistent with the previous report but there are slight differences in their chromosome morphology.*

**Key words**:Hedgehog, Cytogenetics, Karyotype, Micro-chromosome, Diploid, Autosomal Chromosomes.

## Introduction

Hedgehog is the common name for small insectivorous mammals of the family Erinaceidae, which comprises six genera and fifteen species. They are distributed in Africa, Europeand Asia, including certain parts of Indo-Malaysia (Wilson and Reeder, 2005), with some species having wide distribution ranges (He *et al*., 2012).*Atelerix albiventris* also known as the African pygmyhedgehog is a species of hedgehog found throughout much of central and eastern Africa (Santana *et al*., 2010). They inhabit steppes, savannas, grasslands and agricultural fields while trying to avoid forested areas.*A. albiventris* is one of the four members of the genus *Atelerix*. It is the smallest of the African hedgehogs (Santana *et al*., 2010) with increasing popularity as exotic petsand objects of biomedical research (Coker *et al*., 2018). Cytogenetic studies of *A. albiventris* was first reported by Hubner *et al*., 1991 and it is the only existing report on the cytogenetics of this species. Cytogenetic studies are of great significance in revealing the taxonomic identity of a species as well as elucidating the phylogenetic relationships between groups. This study therefore seeks to re-evaluate the cytogenetic characteristics of *Atelerix albiventris* and compare it with the previous report by Haubner*et al*. (1991).

## Materials and Methods

*Collection of Samples*

Four live specimens (two males and two females) of the African Pygmy Hedgehog, *A. albiventris*, were collected from Akesan Market, Oyo town ($7^{o}$46'N, $3^{o}$56'E), Oyo State. External body measurements were taken and recorded for the purpose of identification. Morphological parameters measured were body weight (BW), head length (HL), tail length (TL), hind-limb length (HdL), snout length (StL), body length (BL), body breadth (BB) and teat number (TN) for females. All measurements were in millimeters, using the meter rule. The hedgehogs were identified using the morphological descriptions established by Allen (1992) and the anatomy of the internal sexual organs, in addition to the inspected external genitalia, used to determine the sex of the specimens.

*Chromosomal Analysis*

The sampled hedgehogs were fed and injected intraperitoneally with 0.05% colchicine. Colchicine (prepared with distilled water) was administered at the rate of 1.0ml per 100 grams of body weight. The animals were left for about four to five hours before sacrificing in a killer jar using chloroform. Metaphase chromosomes were prepared from the bone marrow, following the method of Hsu and Patton (1969). The femurs and the humeri were cut out and the bone marrows in them were flushed out with freshly-prepared warm 0.56% potassium chloride solution, using syringes,into centrifuge tubes. The bone marrow solution was checked to ensure no tissues and lipids were present and shaken vigorously, to disperse the cells. The solution was allowed to stand for about 50 minutes.The centrifuge tubes containing the solution were centrifuged at 1000 rpm for 5 minutes. The supernatant was carefully removed with Pasteur pipette, leaving about 0.5ml, in which the cells were re-suspended. A reasonable volume of freshly-prepared Clarke's fluid (ethanol-acetic acid, 3:1) was added, re-centrifuged at 1000 rpm for another 5 minutes and the supernatant removed. The cells were again fixed in two more changes of the fixative and the supernatants removed as in the first fixation. The cells were re-suspended and a freshly prepared ethanol-acetic acid 1:1 fixative was added, to give a reasonable cell concentration for spreading on slides.

The slides to spread on were cooled inside cold distilled water and allowed to dry. The slides were held at an angle of 45°C and the cell suspension dropped from a Pasteur pipette, at a height of about 50 centimeters. The slides were then blown along their lengths to ensure a good spread of the cells. Each slide was labelled and placed on the slide warmer to dry out, at 60°C, prior to staining. Ten slides were prepared for each specimen.

A 6% Giemsa stock solution was prepared by adding 6ml of the stock Giemsa stain to 100ml of the phosphate buffers, comprising 50ml each of disodium hydrogen phosphate ($Na_2HPO_4$) and potassium di-hydrogen phosphate ($KH_2PO_4$). The two buffers were poured simultaneously into the staining jar and the Giemsa added thereafter. The dried slides were stained for about 25 minutes and removed. The stained slides were rinsed with distilled water and allowed to dry on the slide warmer, at 60°C.

*Karyotype Studies*

The slides were viewed under a binocular light microscope to locate metaphase chromosomes. Metaphase chromosomes were then viewed under the scanning power (x40) objective eyepiece and chromosomes numbers were counted. The chromosomes considered good were further

examined using the high power (x100) objective eyepiece under oil immersion and photographed. The morphology of the chromosomes, the diploid chromosome number (2n), the autosomal fundamental number (FNa) and the fundamental number of chromosomal arms were determined by making measurements from the printed photomicrographs of the chromosome spreads.The karyograms were constructed by arranging thechromosomes in pairs of decreasing length and the position of the centromeres was also used in classification.

**Results**

The mitotic metaphase chromosome spread of the male and female*A. albiventris* is shown in Figures 1 and 2 respectively with a diploid number (2n) of 48 chromosomes. The fundamental autosomal number (FN) is 93 in the male and 94 in the female while the autosomal Fundamental Number (FNa) in both male and female *A. albiventris* is 90.A polyploid chromosome spread was also observed in the male*A. albiventris* with 4n = 96 chromosomes. Using the conventional chromosome classification system, both the male and female*A. albiventris* has fourteen pairs of metacentric chromosomes; chromosomes 1, 3, 4, 5, 11, 12, 14, 15, 16, 17, 18, 19, 20 and 21. There are two pairs of submetacentric chromosomes; chromosomes 6 and 13, five pairs of acrocentric chromosomes; chromosomes 7, 8, 9, 10 and 22, and one pair of telocentric chromosomes; chromosome 23 (Fig. 1). The X chromosome is acrocentric in both male and female while the Y chromosome in the male is a micro- chromosome and telocentric. The karyotypes of the male and female*A. albiventris* shows the chromosomes can be grouped into three classes, based on size (Fig 2 and 4). The first class, group A, are the large-sized chromosomes, consisting of three pairs, chromosomes 1, 2 and 3. The second class, group B, are the medium-sized chromosomes,consisting of chromosomes 4 – 19 and theXchromosome. The third class, group C, are the small-sized chromosomes, consisting of chromosomes 20 – 23 and the Y chromosome in male (Fig. 2 and 4). The karyotype observed for the male *A. albiventris* consists of 15 macro-chromosomes and 9 micro-chromosomes, including the Y chromosome (15M + 9m) while that of female *A. albiventris* consists of 16 macro-chromosomes and 8 micro-chromosomes (16M+8m).

**Discussion**

The diploid chromosome number of *A. albiventris,* reported in this study is the same as the one reported in the previous study by Hubner *et al*. (1991). However, there are slight differences in their chromosome morphology as an autosomal fundamental number (FNa ) of 90 and fundamental number of chromosomal arms (FN) of 93 was reported in this study as opposed to (FNa) of 92 and (FN) of 96 that was reported in the previous study (Haubner *et al*., 1991).

Also, in this study, fourteen pairs of metacentric chromosomes; two pairs of submetacentric chromosomes, five pairs of acrocentric chromosomes and one pair of telocentric chromosomes were recorded while in the previous study (Hubner *et al.*, 1991), 13 pairs of metacentric chromosomes, six pairs of submetacentric, two pairs of acrocentric chromosomes and one pair of telocentric chromosomes were recorded.

The diploid chromosome number observed for this species is also the same as that observed in some other species of hedgehogs, including *E. europaeus*(Zima and Kral, 1984), *E. concolor*(Dogramaci and Gunduz, 1993), and *H. auritus* (Arslan*et al*., 2009). It is however, different from

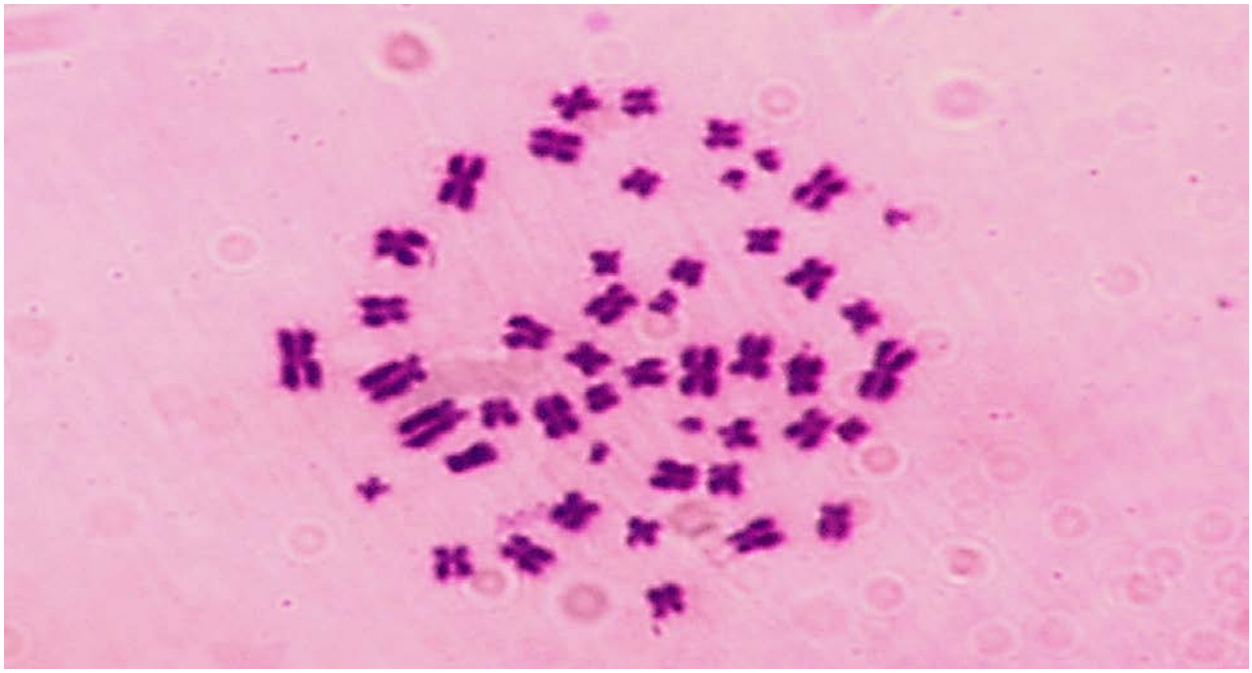

Figure 1: Mitotic metaphase chromosome spread of the male *A. albiventris*

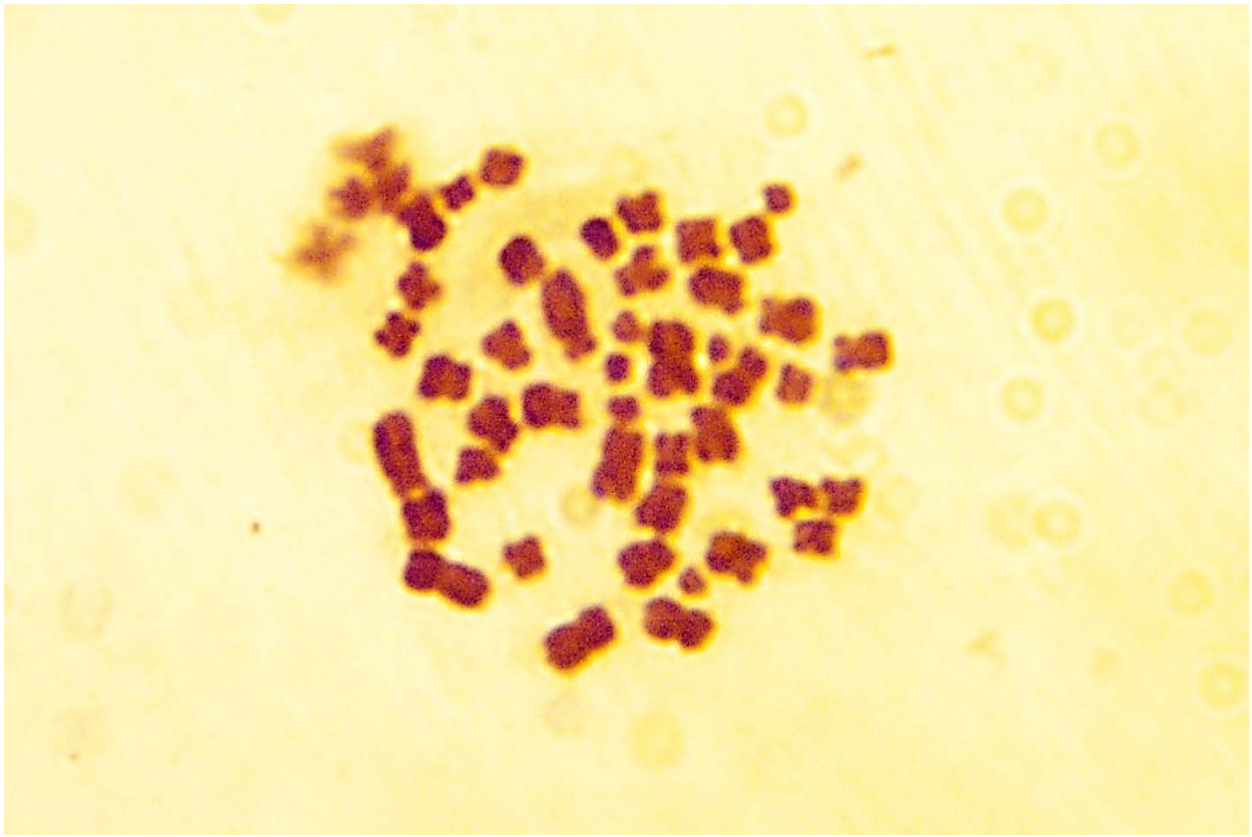

Figure 2: Mitotic metaphase chromosome spread of the female *A. albiventris*

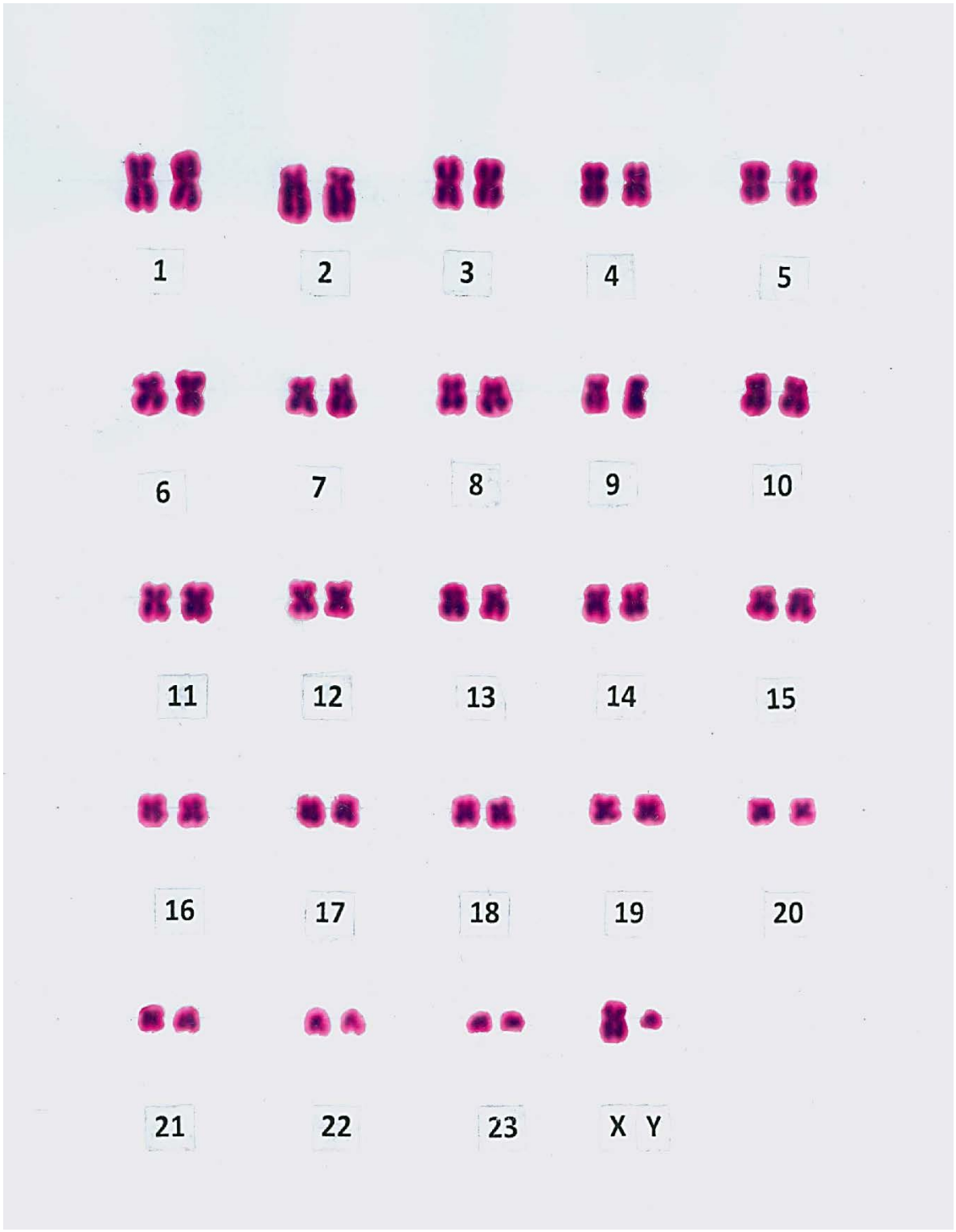


Figure 3: Karyotype of the male*A. albiventris*

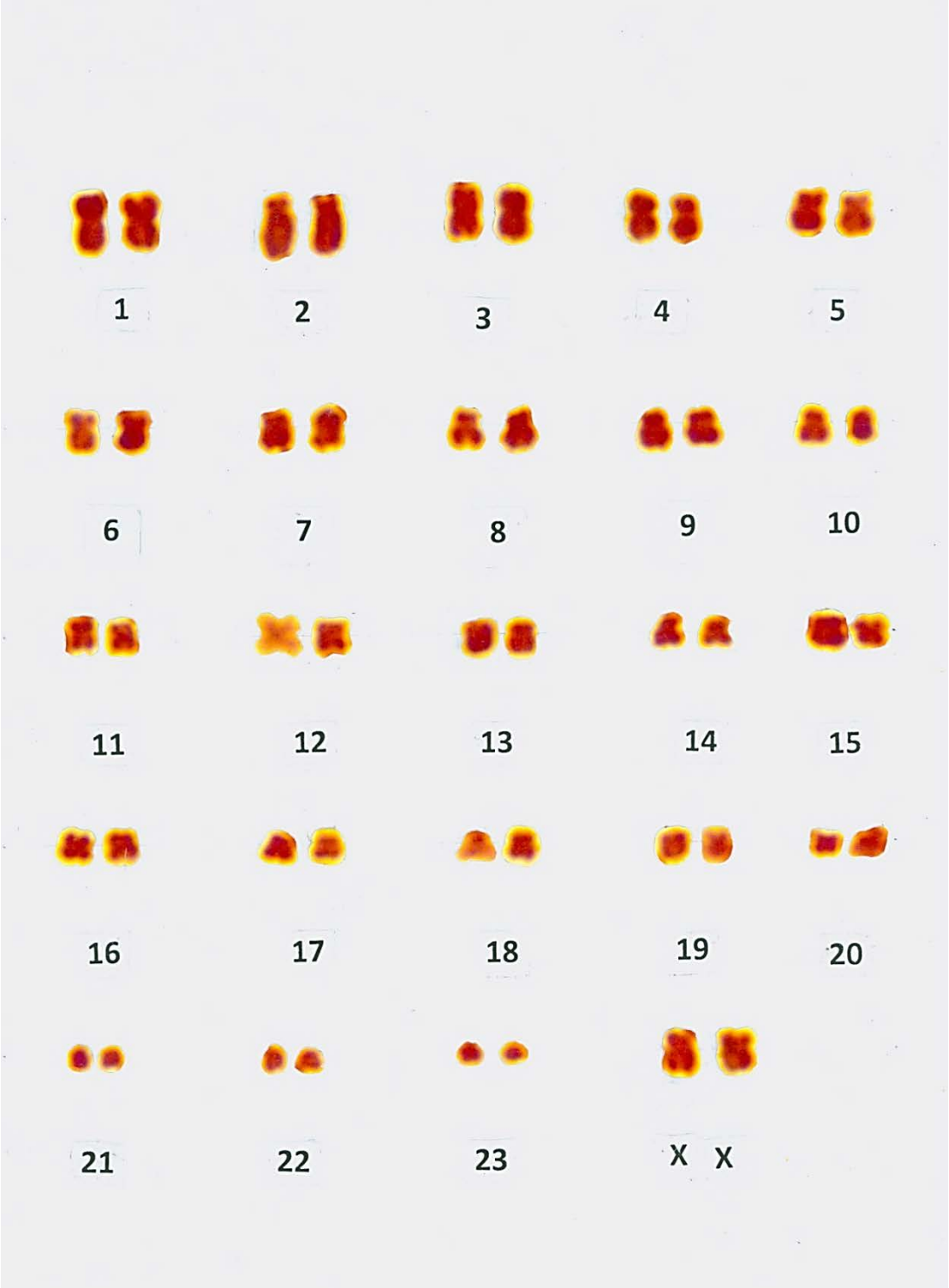


Figure 4: Karyotype of the female *A. albiventris*

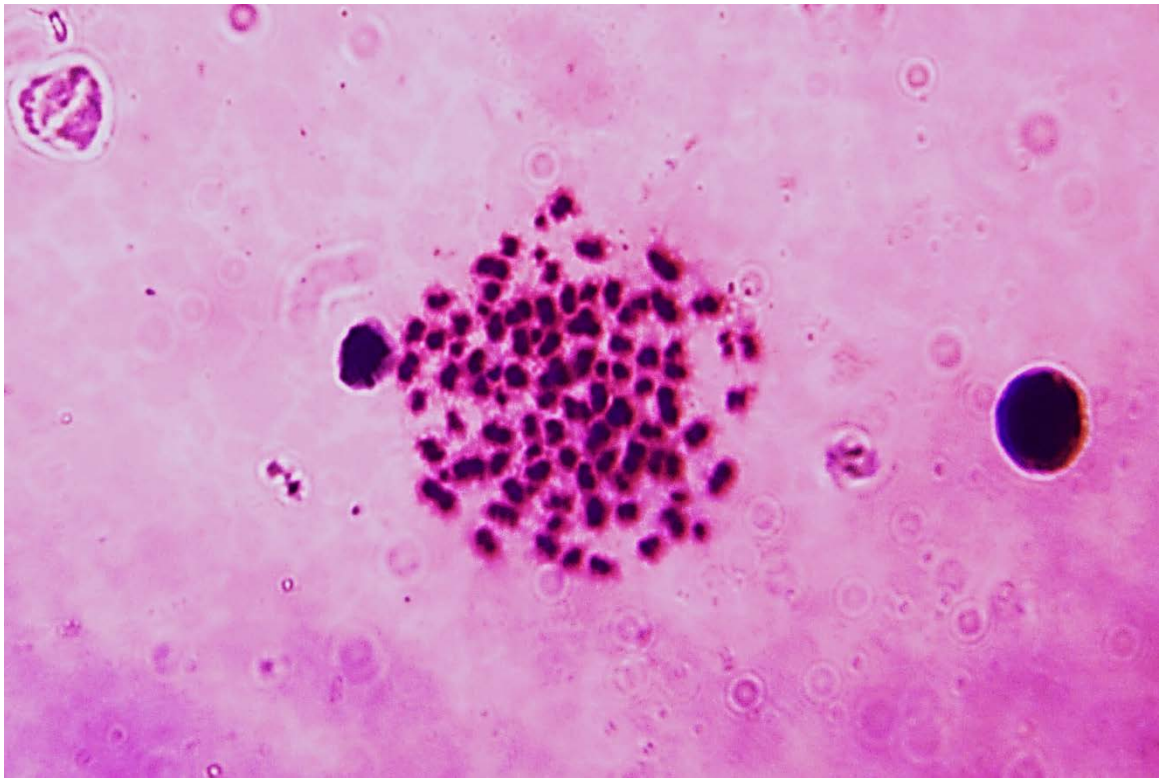

Figure 5: Polyploid spread in the male *A. albiventris*, 4n = 96

that reported for *E. amurensis*, which has a diploid number (2n) of 44 chromosomes (Kang and Kim, 1963). The karyotype observed for this hedgehog species differs when compared across the board with those of the other species. The karyotype for *A. albiventris* in this study records the highest number of acrocentric chromosomes, consisting of six pairs of acrocentric autosomes as opposed to the one pair found in *E. concolor* (Zima and Kral, 1984). No acrocentric chromosomes were reported for the other species previously studied. The X chromosome for *A. albiventris* in this study is a medium-sized acrocentric. The X chromosome is medium metacentric in *E. concolor* (O'Brien *et al*., 2006) and large metacentric in *H. auritus* (Colak *et al*., 1997). The Y chromosome is bi-armed and metacentric in *H. auritus* (Arslan *et al.*, 2009), micro-submetacentric in *E. europaeus* (Hsu and Benirschke, 1967) and minute metacentric in *E. concolor* (O'Brien *et al.*, 2006).

The Fundamental Number of chromosomal arms, FN, observed for *A. albiventris* in this study is 93 for the male and 94 for the female. This differs from that reported for *H. auritus*, which is 96 (Arslan *et al*., 2009) and 94 for *E. concolor*(Dogramaci and Gunduz, 1993). The fundamental number of autosomal arms observed for *A. albiventris* in this study corresponds only with that recorded for *E. concolor*(Dogramaci and Gunduz, 1993). No subtelocentric chromosomes were recorded in this study as have been reported for *H. auritus* (Arslan *et al*., 2009) and *E. concolor* (Dogramaci and Gunduz, 1993).

From the results obtained in this study, it is concluded that the diploid chromosome number of *A. albiventrix* is the same as that reported by Hubner *et al*. (1991). However the slight differences in

their chromosome morphology could have evolutionary/taxonomic implications. Further studies are therefore recommended to check for possible genetic differentiations at the species level.

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