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Karyotypes of two Agamid species from Nigeria

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ABSTRACT: The karyotypes of the common rainbow lizard, Agama agama Lin. and a wild savanna forest relative were investigated, using the bone marrow red blood cells for mitotic chromosomes and the male gametocytes for meiotic chromosomes. The somatic chromosome number of 2n = 44 was confirmed for the rainbow lizard while the relative was found to be 2n = 42. A fundamental number {N.F.} of 46 and 48 respectively were established for them. Chiasma frequency per complement in the two species showed significant difference. High level of polyploidy was observed in the follicular cells among the spermatocytes of both species.

Key words: Karyotype; Agamid; Chromosome morphology; Fundamental number.

Introduction

Lizards are the most abundant of the modern reptiles both in number of kinds and of living individuals. They belong to the Order Squamata of the Class Reptilia. Different species of modern lizards are found in different parts of the world, and they show extensive adaptive radiation which include terrestrial, arboreal, burrowing and aquatic forms (Young, 1981).

The classification of lizards has been found to be problematic and many different systems have been employed. According to bellairs and Attride (1975), the lizards, after originating from the Prolacerta-like ancestors in the early Mesozoic, split into several groups of more or less equal distinctiveness. Based on the assessment of the detailed anatomical structures and cytology, these groups have been assigned the rank of infraorders. These infraorders are Eolacertitia, Gekkota, Iguania, Scincomorpha, Anguimorpha, Platynota, Henophidiaand Caenophidia, and each infraordercontains one or more families.

The Infraorder Iguania consists of three families, viz: Iguanidae, Agamidae and Chamaeleonidae. Members of the family Agamidae include, among others, the genus Agama which consists of both desert living and partly arboreal types. Among the latter is the rainbow lizard, *Agama agama* L. According to Harris (1963), *Agama agama* is widely distributed and it is tolerant to a considerable range of climatic conditions. In Africa, its distribution spans from Ethiopia to Senegal and penetrates southwards into mainland Tanzania and the Republics of the Congo. They inhabit such widely different environments as the humid mangrove swamps and the arid sahara desert. Thiselton (1961) remarked that lizards are the most abundant reptiles in West Africa.

In Nigeria, only two species of the genus *Agama* (*A. agama* L. and *A. bennuensis*) have been identified (Adegoke, per.comm..) While *A. bennuensis* is said to be restricted to the forest zones of the southern part

of the country, *A. agama* is commonly seen in colonies within human habitations throughout the country. Not much has been documented on the cytology and taxonomy of lizards in Nigeria.

The present work is an attempt to record the karyotypes and chromosome number of agamid representatives sampled within the Ilorin metropolis, including the campuses of the University of Ilorin, Nigeria.

Materials and Methods

Specimens of both male and female rainbow lizard were caught during the day time from within the students' hostels of the University of Ilorin, brought into the laboratory and kept in cages or treated immediately for cytological material. Lizards which resembled the rainbow lizard, though smaller in size, were also found in cultivated farmlands around Ilorin. Specimens of this group were also caught and brought into the laboratory.

The rainbow lizard is sexually differentiated both in size and in colour. The mature male is brightly coloured: the head is orange while the trunk is blue and the tail is white, orange and black sectorially. It measures about 15 cm without the tail. The female has dull greenish-brown trunk, dirty white belly while the head has some greenish spots and yellow patches. Pale yellow lines run from the shoulders down to the either sides of the trunk. At maturity, it measures about 10 cm without the tail (Plate 1A and B), this species is gregarious and extremely territorial in habit.

The wild savanna forest species is not sexually differentiated in size and colour. The mature animal resembles the female of the rainbow lizard, though smaller. Both male and female have blue colour on their cheeks and gular folds, with a line extending from the posterior side of the nuchal crest to the tail. They measure about 7 cm at maturity, excluding the tail (Plate 1C). They are more or less solitary. The male is identified by the presence of a row of thick scales (pre-anal pads) at the border of the cloacal opening. Eight animals were sacrificed, two males and two females from each species.

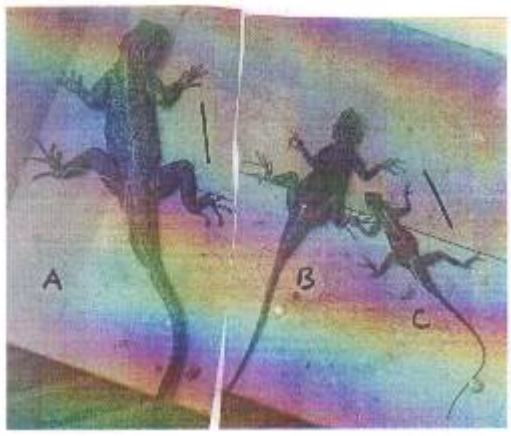

Plate 1: Specimens of the *Agama* lizards.
A: *Agama agama* L. male.
B: *Agama agama* L. female.
C: Wild savanna species, male.
Note: Bar represents 5 cm.

Preparation of cells

Cells for somatic metaphase chromosomes were obtained from bone marrow of the femur and humerus while meiotic cells were obtained from the testes.

The animal was injected with 0.5-1.0 ml of 0.05% colchicines at least two hours before sacrifice. Using a hypodermic needle of appropriate size, attached to a syringe containing 1-1.5 ml hypotonic buffer solution (1% sodium citrate prepared fresh and pre-warmed at 37°C), the marrow was sucked from a dissected bone and aspirated into a 15 ml centrifuge tube, rinsed with 1 ml of the buffer to give a total of 8-10 ml of the material. Fatty material was removed using a Pasteur pipette. Testis was dissected and minced in 1 ml sodium citrate and made up to 8-10 ml with buffer solution in a 15 ml centrifuge tube. In both cases, the cell suspension was shaken and left to stand for 10-15 minutes. The tube was shaken again and centrifuged at 1000 rpm for 5 minutes. The supernatant was carefully sucked with a Pasteur pipette, leaving just 0.3-0.5 ml. 2-2.5ml of freshly prepared fixative (1:3 acetic methanol) was added dropwise with quick shaking after each drop, and centrifuged. The supernatant was again sucked away and the process repeated twice. Cells were resuspended in 2 ml of the fixative and stored in the refridgerator till the next day. The tube was shaken to resuspend the cells and then centrifuged, supernatant reduced to about 0.5 ml and 2-3 ml of freshly prepared acetic methanol added and centrifuged. The supernatant was removed and enough of 1:1 acetic methanol was added to give a reasonable cell concentration on the slide. Cell spread was achieved using the dry spreading method and staining was in 2% Giemsa. Chromosome count was done in at least ten well-spread metaphase plates for each taxon while chromosome analysis and measurements were conducted on at least twenty well-spread metaphase plates. For mitotic chromosomes, measurements were pooled and averages determined for the homologous pairs.

Results and Discussion

The somatic complements of both species contain both macro- and micro-chromosomes. The complement of the rainbow lizard consists of 24 macrochromosomes and 20 microchromosomes in both sexes, giving the species a diploid number of 2n = 44. The wild savanna species, on the other hand, has a complement of 2n = 42, made up of 20 macrochromosomes and 22 microchromosomes. In both species, the macrochromosomes vary in length from over 1.0 µm while the microchromosomes are all less than 1.0 µm. The macrochromosomes are resolvable into morphological pairs in both species. Table 1 contains a summary of the measurements of the macrochromosomes of both species. The longest pair in both species is distinct, being much longer than the others. In the rainbow lizard, this longest pair has a submedianly located centromere while all others are telocentric. In the wild savanna species, the longest and the third pairs both have submedianly located centromere while the second longest pair has subterminally located centromere; all others are telocentric. Hence, karyotypically, the wild species differs from the rainbow lizard. However, the total haploid chromatin length of the macrochromosomes in both species is 35.1 µm. The fundamental number (number of chromosomes arms per somatic complement) is 46 in the rainbow lizard but 48 in the savanna species.

Meiosis was found to be regular in both species. Chiasma frequency was found to be much higher in the rainbow lizard than in the savanna species. Polyploid cells were observed among the follicular cells in both species. Level of ploidy ranged from tetraploidy to 16-ploidy with an incidence of 8-14% in the rainbow lizard and 1-6% in the savanna species.

The karyotype of the Class Reptilia had been reported to be characterized by the presence of small chromosomes referred to as microchromosomes. Although the term is not clear, as remarked by Tegelstrom and Rhyttman (1981), and its definition may be subjective, Ohno (1970) classified chromosomessmaller than 1,.0 µm at full mitotic metaphase as microchromosomes. However, in the family Agamidae as well as Iguanidae, there is a sharp distinction between the macro- and the micro—chromosomes (DeSmet, 1981; White, 1973) which was found to be true in the animals treated here.

The somatic chromosome number of 2n = 44 for the rainbow lizard in this work agrees with the findings of Gorman and Schochat (1972) and Adegoke (per.comm..). It, however, differs considerably from that of de Smet (1981) but the karyotype as reported here corresponds with that of *Agama atra* DAUD as reported by De Smet (1981) in chromosome number, morphology and structure. The variation in chromosome size could be as a result of differences in condensation. On the other hand, the chromosome

number of 2n = 42 for the wild savanna species has not been reported for any known species of Agama in tropical Africa though it falls within the range of the somatic chromosome numbers recorded for the genus as reported by Gorman and Shochat (1972).

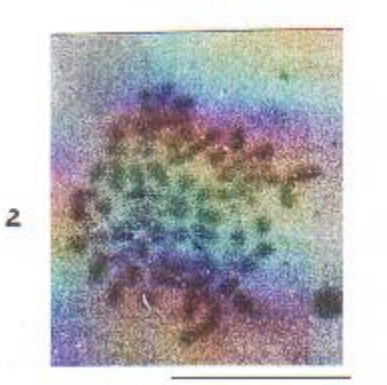

Plate 2: Polyploid (4n) metaphase spread from the spermatocyte of the wild savanna species.

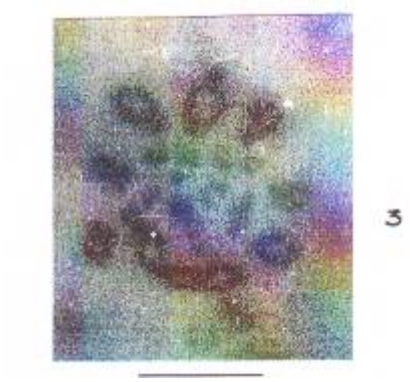

Plate 3: Meiotic metaphase I complement of the male of wild savanna species.

The chromosome size distribution within the somatic complement of both species is characteristic of the lizards (Bull, 1978; Cole, 1979; De Smet, 1981). There are, however, notable differences in the morphology of the chromosomes of the rainbow lizard and its wild savanna relative. Karyotype evolution in the two species is more probably by mere centric fission/fusion. Meiosis in both species is regular, showing that the genetic systems representing both species are stable. While expecting more information on the biology of these animals, it is safe to regard the two as distinct species of the genus Agama, separated by chromosome morphology and karyotype as well as by animal size, ecological preference and ecological behaviour.

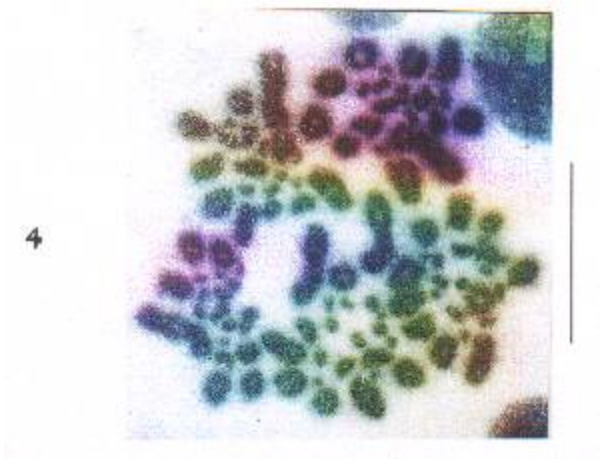

Plate 4: Meiotic metaphase I complements (5 cells) of the male of the male *A. agama* L.

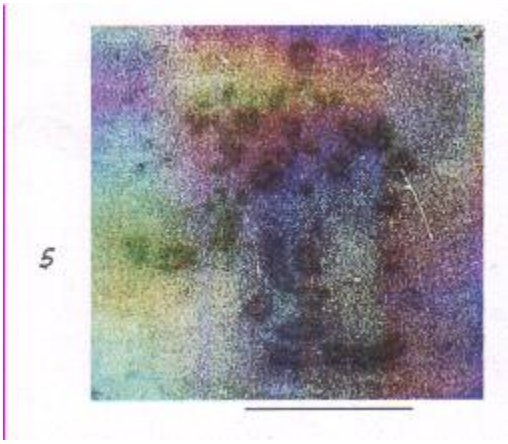

Fig. 5: Somatic complement of male wild savanna species.

Table 1:

Table 1: Summary of Macrochromosome Measurement at Metaphase (in μm).

TAXA		HOMO 1	LOGOUS 2	CHRO 3	MOSOME 4	PAIRS 5	6	7	8	9	10	11	12	Ex
	Long arm	6.35	4.0	3.6	3.3	3.0	2.6	2.5	2.0	1.7	1.3	1.3	1.2	
	Shortarm	2.25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
A	R (l/s)	2.83	0	0	0	0	0	0	0	0	0	0	0	35.1
	Centr. Loc	sm	T	T	T	T	T	T	T	T	T	T	T	
	Long arm	5.6	3.2	2.8	3.5	3.2	3.1	2.6	2.6	2.2	2.0			
	Short arm	2.1	1.0	1.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0			35.1
B	R (l/s)	2.67	3.2	2.3	0	0	0	0	0	0	0			
	Centr. Loc.	sm	st	sm	T	T	T	T	T	T	T			

Note: Taxon A – Agama agama L.
Taxon B – The wild savanna forest species.

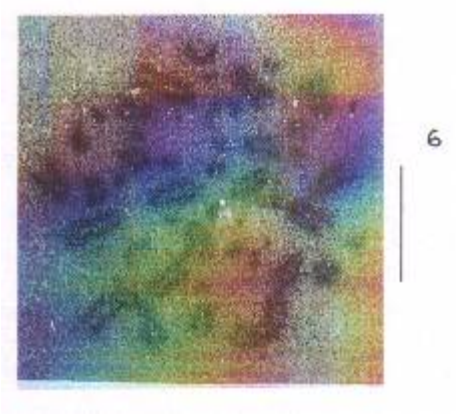

Fig. 6: Somatic complement of male *Agama agama* L.

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