Volume : 2 | Issue : 9 | Sept 2013 • ISSN No 2277 - 8160

Research Paper

Biotechnology

Genetic Surveillance of Kingfisher & Bee Eater

H. K. Garg — Department of Zoology & Biotechnology, Government Motilal Vigyan Mahavidyalaya, Bhopal - 462008, India

Ashish Shrivastava — Department of Biotechnology, CSA Government Post Graduate College, Sehore - 466001, India

ABSTRACT

The present study deals with the complete morphometric analysis of chromosomes of two coraciiform birds - king fisher, Ceryle rudis and green bee-eater, Merops orientalis. The chromosomes were extorted from marrow-cells of previously colchicinized grown-up individuals. The chromosome tally varied between 78 to 84 in Ceryle rudis and 80 to 85 in Merops orientalis with a prominent peak at 82 in both the species. There was no distinction in size between macro- and micro- chromosomes

KEYWORDS:

Introduction

There are 8,948 species of living birds. Of this, 802 have been karyotyped so far, which serve chromosomal information for less than 8% of the global bird fauna (Garg & Garg, 2003; Garg & Shrivastava, 2013). In spite of forthcoming danger for innumerable bird species to join the roll of 'endangered' or 'threatened' forms, we barely know anything about their cytogenetic framework. In order to explore the hereditary composition of some new avian taxa and to validate their taxonomical resemblance, sixteen specimens of king fishers and bee eaters have been subjected to karyological assessment.

Material & Methods

Five male and two female specimens were procured from different parts of the state. The individuals were anaesthetized at the site itself and their bone marrow, extracted through a sternal puncture, were brought to the laboratory for further treatment. The cell plates were prepared after Rothfels & Siminovitch (1958) with certain revisions. The chromosomes were classified on the basis of Percentage Relative Length (% $_{L}^{R}$) and Arm Ratio (r) as follows :

$$\text{Percentage Relative Length } (\%\ {}_{L}^{R}) = \frac{\text{Length of any macrochromosome}}{\text{Total macrochromosomal length}} \times 100$$

$$\text{Arm ratio (r)} = \frac{\text{Length of long arm}}{\text{Length of short arm}}$$

as given by Levan *et.al.* (1964).

Results

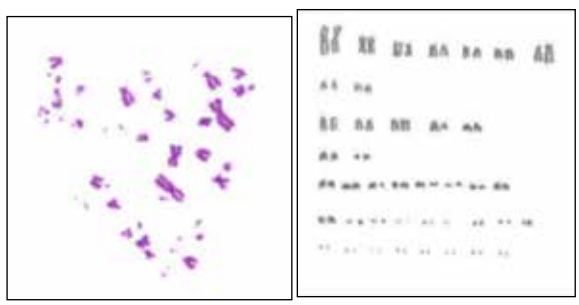
King fisher, Ceryle rudis

The bone marrow cells of five male and two female specimens yielded one hundred thirty six metaphase plates. The diploid number of chromosomes for the species was construed to be 82 with a variation range from 78 to 84.

Unlike general condition in birds, there was hardly any discrimination between the macro- and micro- chromosomes, in this species. However, following the archetype offered by De Boer (1976), first sixteen pairs of chromosomes, sharing ≥ 2.5% of the total chromosomal length, have been classified as macrochromosome.

The macrochromosomes were arranged, as per homologies based on the criterion of percentage relative length as well as centromeric index, into four groups :

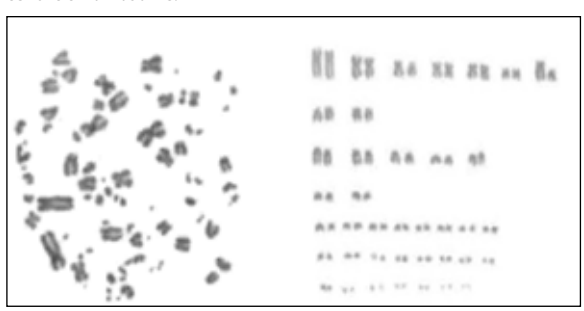
Group I was made of six biarmed metacentric pairs of autosomes (chromosomes 1-6). Chromosome-1, was the largest element of the karyotype with a mean absolute length of 5.26µ, constituted 12.54% of the TML. Chromosome - 2 ranked subsequently in the order of size and had a higher centromeric index than chromosome - 1 (chromosome - 1 : $_{I}^{c}$ = 39.53 ; chromosome - 2 : $_{I}^{c}$ = 44.73). The chromosome 3, 4, 5 and 6 showed a continuous gradation in size ($_{L}^{R}$ = 7.02%, 6.41%, 6.01% 4.22% respectively) and it was rather difficult to individualize them.



Metaphase plate & karyotype of *Ceryl rudis* (male)

Group II covered two pairs of autosomes (chromosomes 7 & 8) and a Z chromosome. Chromosome - 7 and 8 were, nevertheless, larger in size than chromosome - 6 ($_{L}^{R}$ = chromosome - 6 : 4.22% ; chromosome - 7 : 5.60% ; chromo some - 8 : 5.11%).

However, there was a fair reduction in centromeric indices among the three pairs ($_{I}^{c}$ = chromosome - 6 : 44.54 ; chromosome - 7 : 35.48 ; chromosome - 8 : 29.54). The sex element - Z was a large sized sub-metacentric chromosome.



Metaphase plate & karyotype of *Ceryl rudis* (female)

Morphometric data of macrochromosomes has been given hereunder :

Chromosome Number	% Relative Length	Centromeric Index	Chromosome Type
1	12.54 ± 0.16	70.00 ± 0.72	m
2	09.63 ± 0.05	42.64 ± 0.81	m
3	07.02 ± 0.11	40.04 ± 0.44	m
4	06.41 ± 0.07	39.11 ± 0.63	m
5	06.01 ± 0.88	38.17 ± 0.66	m
6	04.22 ± 0.51	44.54 ± 0.46	m

7	05.60 ± 0.63	35.48 ± 0.99	sm
8	05.11 ± 0.12	29.54 ± 0.30	sm
9	07.46 ± 0.66	24.21 ± 0.87	st
10	05.44 ± 0.87	25.03 ± 0.34	st
11	05.17 ± 0.52	24.17 ± 0.88	st
12	04.96 ± 0.39	20.05 ± 0.17	st
13	04.44 ± 0.41	19.18 ± 0.12	st
14	03.82 ± 0.64	-	T
15	03.04 ± 0.14	-	T
Z	09.13 ± 0.57	33.33 ± 0.66	sm
W	03.98 ± 0.77	45.00 ± 0.71	m

Group III included five pairs of sub-telocentric chromosomes, (chromosome 9 - 13) with overlapping range of relative length and centromeric indices.

Group IV was represented by two small sized telomeric pairs, chromosomes - 14 and 15, depicting similar morphlogy. These pairs were even smaller than the W chromosome ($_{L}^{R}$ = chromosome - 14 : 3.82% ; chromosome - 15 : 3.04% ; W - chromosome : 3.98%).

A total number of twenty five dot shaped pairs, other than sixteen described so far, formed a continuous series and were placed together as microchromosomes.

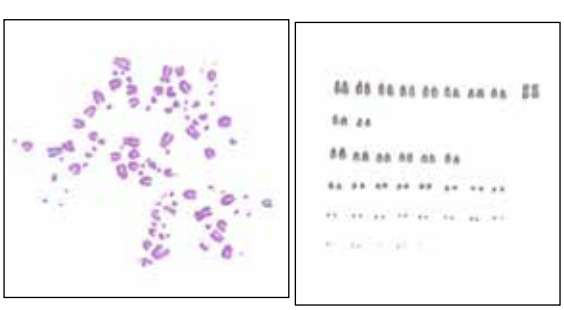
Green bee-eater, Merops orientalis

Four male and five female specimens of green bee-eater were collected during suitable season. Of some seventy-plus metaphase plates examined, thirty cells had 41 pairs of chromosomes (2n=82). Although in rest of the cells, the diploid chromosome count varied from 80 to 85.

On arranging the chromosomes into morphologically similar pairs, no significant variation was noticeable within the chromosomes. There was a steady decline in size, so much so, that it was difficult to demarcate the chromosomes into two conventional categories. However, applying the notion given by De Boer (1976), seventeen pairs of chromosomes contributing ≥ 2.5% of total chromosomal length had been differentiated as macrochromosomes.

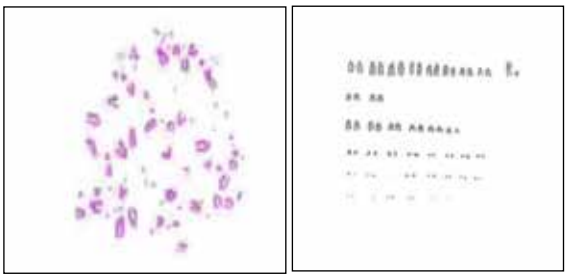
On the basis of nomenclature proposed by Levan *et al.* (1964), the macrochromosome were classified into four groups. Group I was represented by a sex chromosome (Z). It was the largest element of the karyotype ($_{L}^{R}$ = 9.54% and $_{I}^{C}$ = 35.28).

Group II comprised two pairs of large (≥ 7.5% of TML) and six pairs of medium (≥ 2.5% of TML) sized subtelocentric chromosomes (chromosome 1 - 8). e of them was easily separable owing to a regular fall in size, comparable morphology and overlapping array of centromeric indices.



Metaphase plate & karyotype of *Merops orientalis* **(male)**

Group III consisted of two pairs of autosomes (chromosome - 9 & 10) with terminally positioned centromeres.



Metaphase plate & karyotype of *Merops orientalis* **(female)**

Group IV included six pairs of intermediate sized chromosomes (chromosome 11 - 16). They had a terminal centromere (*sensu stricto*) adding together 25.78% of TML.

W, a metacentric chromosome, constituted 3.99% of TML.

Morphometric data of macrochromosomes has been given hereunder :

Chromosome Number	% Relative Length	Centromeric Index	Chromosome Type
1	8.48 ± 0.26	22.20 ± 0.42	st
2	7.60 ± 0.45	18.74 ± 0.67	st
3	7.44 ± 0.11	17.88 ± 0.32	st
4	7.06 ± 0.21	16.54 ± 0.92	st
5	6.61 ± 0.86	17.98 ± 0.74	st
6	6.11 ± 0.34	18.01 ± 0.61	st
7	5.88 ± 0.23	17.77 ± 0.99	st
8	5.76 ± 0.41	21.01 ± 0.83	st
9	5.00 ± 0.72	12.50 ± 0.78	t
10	4.74 ± 0.33	12.29 ± 0.14	t
11	5.03 ± 0.12	-	T
12	4.80 ± 0.09	-	T
13	4.36 ± 0.81	-	T
14	4.03 ± 0.76	-	T
15	4.00 ± 0.42	-	T
16	3.56 ± 0.61	-	T
Z	9.54 ± 0.49	48.83 ± 0.50	m
W	3.99 ± 0.61	35.28 ± 0.33	sm

Apart from these seventeen pairs, there were twenty four small dot-shaped telocentric elements, scattered hither and thither in the cell plates, amounted to 24.11% of the total genome.

Discussion

Coraciiformes, represented by kingfishers and bee eaters, is known to be the most heterogenous assemblage among avian orders (2n ranges from 68 to 126). So far two species of kingfishers viz. *Dacelo gigas* and *Halcyon smyrnensis* have been karyotyped (Bhunya & Mohanty, 1987 a). They share gross karyological uniformity characterized by the presence of an exceptionally large chromosome-1, almost triple in size than any other chromosome in the genome. On the contrary, no similar chromosome could be seen in *C. rudis* (present study). It supports the argument of Sibley *et al.* (1988) who assigned an individual familial rank to each species.

Coraciiformes, represented by kingfishers and bee eaters, is known to

be the most heterogenous assemblage among avian orders (2n ranges from 68 to 126). So far, two species of kingfishers viz. *Dacelo gigas* and *Halcyon smyrnensis* have been karyotyped (Bhunya & Mohanty, 1987). They share gross karyological uniformity characterized by the presence of an exceptionally large chromosome – 1, almost triple in size than any other chromosome in the genome. On the other contrary, no similar chromosome could be seen in *C. rudis* (present study). It supports the argument of Sibley *et al. (*1988) who assigned an individual familial rank to each species.

In contrast to kingfishers, bee-eaters viz. *M. philippinus* and *M. orientalis* were found to share similar karyological features. The genomes, in both the species, were devoid of large autosomes. The sex-element (Z - chromosome) was the largest member of the karyotype, contributing 9.5 ± % of TML.

However, there is lack of consensus regarding the exact diploid number and mechanism of sex-discrimination. During present study, a total number of 82 chromosomes (41 pairs) were reckoned in atleast 30 out of 72 metaphase plates. *Au contraire,* Sultana & Bhunya (1987) reported 2n to be 78 for females and 79 for males. According to them, a telocentric C-band positive chromosome exists in male as well as female karyotypes of *M. orientalis* and they construed it as a W - chromosome. It was inferred to be a subsequence of non-disjunction of Z-chromosome during meiosis that might have resulted in the formation of sperms with 38 autosomes + ZZ chromosomes. Such sperms, on fertilization with normal eggs (38 autosomes + W chromosomes), produced a male offspring with 76A + ZZW and a female progeny with 76A + ZW constitution. No such element, corresponding to the unpaired submetacentric chromosome, branded as 'W' in females, was discernible in male cell-plates.

In order to verify the presence of W in male cell-plates, certain other techniques like avian leucocytes' culture, feather pulp (in vitro), and banding pattern of chromosomes were attempted but for lack of classy facilities like Nikon Optiphot Zeiss/Lietz with photography phone fluorescence.

From the karyotype of king fishers and bee-eaters, it is evident that they have different shapes, morphology and number of macrochromosomes. Even, the chromosome - 1, which is considered to be highly conserved in avian orders, varied significantly in kingfishers and bee-eaters.

REFERENCES

| BHUNYA S.P. & MOHANTY M.K. (1987) : Studies of the karyotypes and bandings of 41 species of Indian birds (Aves). Chrom. Inf. Serv., 42 : 12-15. | | DE BOER L.E.M. (1976) : The somatic chromosome complements of sixteen species of Falconiformes (Aves) and the karyological relationships of the order. Genetica, 46 : 77-113. | | GARG H.K. & GARG J. (2003) : Chromosomal aberration in a piciform bird, Megalaima zeylanica caniceps (Franklin). Ind. J. Appl. Pure Biol., 18 (2) : 135-140. | | GARG H.K. & SHRIVASTAVA (2013) : Genetic organization of Motacilla flava. International Journal of Scientific & Technology Research – 0913-7193 (accepted for publication). | | LEVAN A., FREDGA K. & SANDBERG A.A. (1964) : Nomenclature for centromeric position on chromosomes. Hereditas, 52 : 201-220. | | ROTHFELS K.H. & SIMINOVITCH C. (1958) : An air drying technique for flattening chromosomes in mammalian cells grown in vitro. Stain Tech., 33 : 73-77. | | SIBLEY G.G., AHLQUIST J.E., BURT L.M. Jr. : A Classification of the living birds of the world based on DNA-DNA Hybridization studies. The Auk., 105, 409-423 (1988). | | SULTANA T. & BHUNYA S.P. (1987) : An unusual chromosomal constitution in male Merops orientalis Latham (Meropidae : Coraciformes : Aves). The Nucleus, 30 : 40-44. | . |