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Chromosome study of *Amazona amazonica* and *A. aestiva* (Aves: Psittaciformes): determination of chromosome number and identification of sex chromosomes by C-banding methods

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Abstract

Two parrot species, *Amazona amazonica* and *A. aestiva*, submitted to cytogenetic analysis presented a diploid chromosome number of 2n=70 (20M + 50m). With the C-banding pattern, the cells of female specimens showed an almost totally heterochromatic W chromosome. No chromosome differences were observed in the two species studied.

Introduction

Amazona amazonica is found in the Amazon region, occurring also in the states of Mato Grosso, western São Paulo, Rio de Janeiro and Paraná, in Brazil (Sick, 1985). They are typically coastal birds feeding on fruits growing in these regions. They live rigorously in couples, keeping social contact with the flock (Forshaw, 1973). *Amazona aestiva* lives in the forest, in palm tree woods and along river banks. They are geographically distributed throughout north-eastern Brazil (Piauí, Pernambuco and Bahia), central Brazil (Minas Gerais and Mato Grosso) and the southern part of Rio Grande do Sul (Sick, 1985). They feed on fruits, grains, seeds, nuts, flowers and leaf sprouts. They live in couples or in small flocks (Forshaw, 1973).

Many parrots have no external sexual dimorphism. An adequate method for sexing parrots is by means of a chromosome study. The sex determining mechanism in birds is of the ZZ/ZW type. The genetic identification of the sex is relatively easily performed due to the relatively large size of the Z and small size of the W chromosome. The size of the Z chromosome is practically identical for different species. On the other hand, the W chromosome varies widely in size. In most birds the W is almost entirely heterochromatic.

Material and methods

Amazona amazonica and *A. aestiva* were obtained from private breeders in Ribeirão Preto, SP, Brazil.

Cytological preparations were made by the direct method with feather pulp material. The Ferrari method utilized (Giannoni *et al.*, 1986) is as follows: young feathers with well-developed pulp are placed in a hypotonic solution of 0.075M KCl plus 6.4 g/ml colchicine for about 1 hour to separate the pulp. After centrifugation and removal of the supernatant, the material is fixed with methanol: acetic acid (3:1). Slides are prepared with 1 to 3 drops of cell suspension and stained with Giemsa.

The G-banding pattern was obtained using Scheres' method (1972) with some modifications.

The intensive staining of the W sex chromosome through the C-banding pattern was obtained by the Summer (1972) technique with modifications.

Results

Figure 1 shows the karyotype of an *Amazona amazonica* female. The most frequent diploid chromosome number was found to be 70.

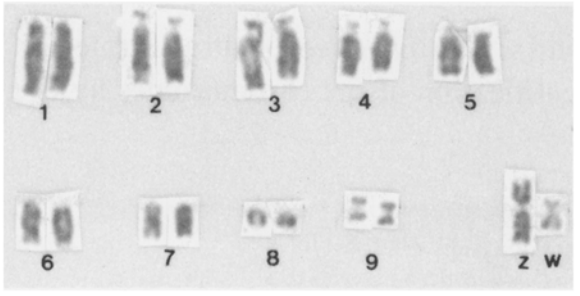

Fig. 1. Partial karyotype of a female of *Amazona amazonica.*

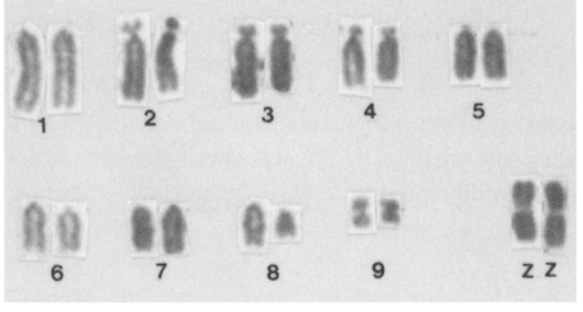

Fig. 2. Partial karyotype of a male of *Amazona aestiva.*

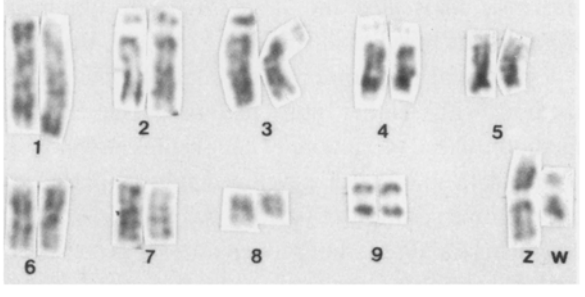

Fig. 3. G-banding pattern of a female of *Amazona amazonica.*

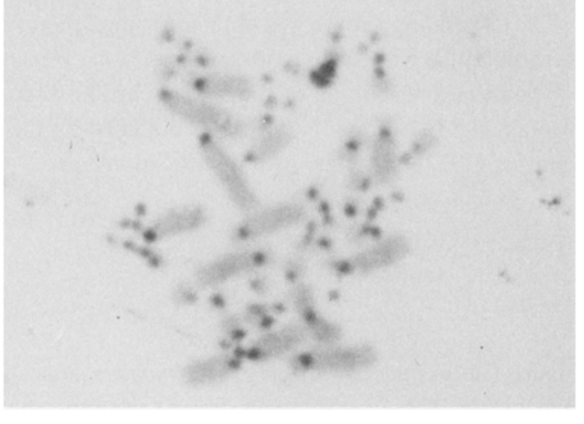

Fig. 4. C-banding pattern of a female of *Amazona amazonica.*

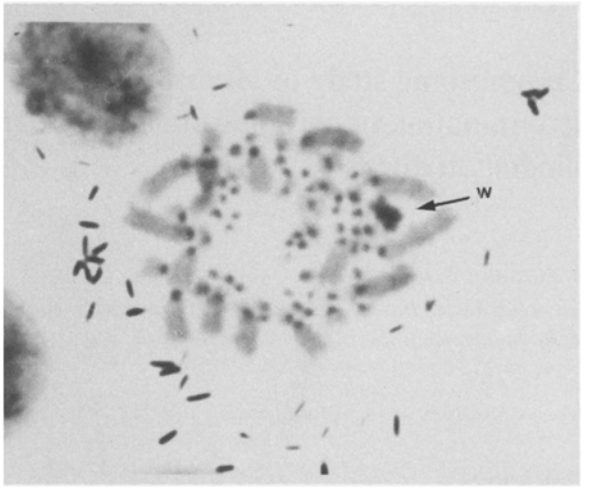

Fig. 5. C-banding pattern of a female of *Amazona aestiva.*

Figure 2 shows the karyotype of an *A. aestiva* male. The most frequent diploid chromosome number was found to be 70.

The number and morphology of the chromosomes of both species were practically identical. The first ten chromosome pairs corresponded to the macrochromosomes and the remaining ones were considered to be microchromosomes, some of which were identified as being two-armed. The first pair consisted of clearly distinct large acrocentric chromosome. Pairs 6, 7 and 8 were also acrocentric. Pairs 2, 3, 4 and 5 were subtelocentric. The ninth pair and the sex chromosomes Z and W were metacentric. The transition from macro to microchromosome was quite distinct.

Figure 3 shows the G-banding pattern of *A. amazonica* macrochromosomes.

Figures 4 and 5 show the C-banding pattern of metaphase chromosomes of an *A. amazonica* and *A. aestiva* female, respectively, where we can clearly observe the presence of a deeply stained W sex chromosome. The C-banding pattern of the two species were similar.

Discussion

The *Amazona amazonica* karyotype described here for the first time, and the *A. aestiva* karyotype studied in this paper present a diploid chromosome number of 70. They have a relatively large number of telocentric chromosomes as compared with the other species of

Psittacidae. Within *Amazona*, only three species had been previously studied, the yellow-crowned Amazon (*A. ochrocephala*) with 2n=70 (De Boer & Belterman, 1980), the blunt-tailed Amazon (*A. viridigenalis*) with 2n=72 (Van Dongen & De Boer, 1984) and the blue-fronted Amazon (*A. aestiva*) with 2n-68 (Schmutz & Prus, 1987). All Amazon species studied have similar macrochromosomes and sex chromosome pairs. There is no clear boundary between macro- and microchromosomes in *A. ochrocephala*, but we found such a distinction in *A. amazonica* and *A. aestiva*.

The karyotypes of *A. ochrocephala* and *A. viridigenalis* are very much alike. The only difference concerns the slightly larger number of microchromosomes in the latter species. The karyotypes of *A. amazonica* and *A. aestiva* are identical and very similar to those of the two *Amazona* species reported in the literature. They have a large acrocentric pair 1; pair 2, 3 and 4 are subtelocentric, and pairs 5, 6, 7 and 8 (only present in *A. amazonica* and *A. aestiva*) are acrocentrics of medium size. Pair 8 in *A. viridigenalis* and *A. ochrocephala* and pair 9 in *A. amazonica* and *A. aestiva* are the only small metacentric elements in the karyotype. The Z chromosome is metacentric and about the size of pair 5. The W chromosome is metacentric as well and somewhat larger than the chromosomes of pair 8. On the other hand, the *A. aestiva* karyotype previously described by Schmutz and Prus (1987) shows numerical and morphological differences when compared to the one presented in this paper. The karyotype described by these autors lacks the large acrocentric pair 1, the medium-sized acrocentric pair 8 and the large subtelocentrics pairs 2, 3 and 4 observed in our study.

The G-banding pattern of *A. amazonica* shown here for the first time was very difficult to obtain.

The C-banding patterns of *A. amazonica* and *A. aestiva* have not been studied previously. Chromosome bands are relatively difficult to obtain in birds. De Boer (1984) reported that out of the 587 species of birds studied, the karyotypes of only 81 were analysed by using chromosome banding pattern techniques. Cytogenetic studies are important to clarify doubts about the taxonomy of several groups of vertebrates, as well as for the sexing of animals without sex dimorphism. Most Psittaciformes present sex chromosomes in the 5th position and varying from submetacentric to metacentric in morphology. The species of the genus *Amazona* follow this general characteristic. Both *Amazona* species studied here present metacentric Z and W sex chromosomes in the 5th position. Determination of the sex chromosome pair by C-banding is a procedure suitable for sexing Psittaciformes. The W chromosome is the most heterochromatic of the pair (Figs. 4 and 5).

Acknowledgements

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