Brazilian Journal of Genetics, 18, 4, 623-628 (1995)
(Revista Brasileira de Genética)

SHORT COMMUNICATION:

Cytotaxonomic analysis of Brazilian species of the genus *Amazona* (Psittacidae, Aves) and confirmation of the genus *Salvatoria* (Ribeiro, 1920)

José Maurício Barbanti Duarte and Renato Caparroz

ABSTRACT

The karyotypes of 12 species of Psittacidae of the genus *Amazona* were studied: *A. aestiva, A. amazonica, A. brasiliensis, A. autumnalis, A. farinosa, A. festiva, A. kawalli, A. ochrocephala, A. pretrei, A. rhodocorytha, A. vinacea* and *A. xanthops*. The metaphases were obtained using a short term culture of feather pulp. Eleven of the twelve analyzed species were karyotypically homogeneous, with only a few divergences in chromosomes 2 and 3. The species *A. xanthops* showed large karyotypic differences compared to the genus *Amazona*. Consequently, the genus *Salvatoria* (Ribeiro, *Rev. Mus. Paul. 12*: 1-82, 1920) was confirmed, and *A. xanthops* renamed *Salvatoria xanthops*.
The study showed the chromosomic conservation of the genus *Amazona* and the need for further taxonomic studies of the karyotype of the Psittacidae.

INTRODUCTION

Cytogenetic techniques have been used recently in the evolutionary and taxonomic study of wild animals. In birds, the feather has been the material most used to get metaphases. The method was introduced by Sandnes (1954) and modified by Shoffner *et al.* (1967). Other authors have worked with blood (Sasaki *et al.*, 1984) or fibroblast cultures (Sasaki *et al.*, 1968; Jovanovic and Atkins, 1969).

There were eleven species of Psittacidae of the *Amazona* genus described for Brazil (Sick, 1984) and, recently, another species, *A. kawalli* (Grantsau and Camargo, 1989), was identified.

Departamento de Melhoramento Genético Animal, Faculdade de Ciências Agrárias e Veterinárias, UNESP, 14870-000 Jaboticabal, SP, Brasil. Send correspondence to J.M.B.D.

Four of the five species of the *Amazona* genus already submitted to cytogenetic analysis are found in Brazil. The basic karyotype of the species varies little, with the diploid number varying from 70 in *A. ochrocephala* (Boer and Belterman, 1981), *A. amazonica* (Aquino and Ferrari, 1990) and *A. aestiva* (Schmutz and Prus, 1987; Aquino and Ferrari, 1990) to 72 in *A. autumnalis* (Lucca *et al.*, 1991) and *A. viridigenalis* (Van Dongen and Boer, 1984). Other differences are based on chromosome classification and organization. Lucca *et al.* (1991) considered the large telocentric chromosomic pair as the third, and not the first pair, as do most authors. Aquino and Ferrari (1990) considered 10 pairs of macrochromosomes whereas others considered only nine. In *A. ochrocephala*, Boer and Belterman (1981) described the eighth chromosome pair as the fifth pair. Falcone (1991), working with *A. aestiva* and *A.*

amazonica, numbered the two-arm chromosomes initially, followed by the telocentric pairs.

In spite of these differences, the karyotypic analysis has demonstrated a strong similarity among the various species of the *Amazona* genus, suggesting great chromosomic conservation in this group.

MATERIAL AND METHODS

A total of 114 birds of the *Amazona* genus were used in this study, being 42 of *A. aestiva*, 10 of *A. amazonica*, 17 of *A. brasiliensis*, seven of *A. farinosa*, two of *A. autumnalis*, seven of *A. festiva*, one of *A. kawalli*, five of *A. ochrocephala*, 10 of *A. rhodocorytha*, seven of *A. vinacea*, four of *A. xanthops* and two of *A. pretrei*.

The metaphases were obtained using a short term culture of growing feather pulp technique. Five feathers, approximately 25 days old, were collected from the high dorsal region, between the neck and the wing. The feathers were cleaned externally with 70% alcohol and squeezed from the tip to the base to extract the gelatinous bulb from the inside of the feather. The bulb was placed in a complete culture medium (8.0 ml Mc Coy's, 1.0 ml of inactivated horse serum and 0.6 ml of phytohemagglutinin). The material was incubated at 39°C for six hours and exposed to colchicine (three drops of 0.0016% solution) in the last two hours of incubation. Centrifugation was used to remove the culture medium and the remaining material was hypotonized in a 0.075 M solution of KCl for 15 minutes at 39°C. The rest of the technique followed the usual standards of fixing, slide making, staining and analysis.

Chromosomal biometry was used for the determination of the chromosome arms ratio and classification according to Levan *et al.* (1964).

RESULTS

The diploid chromosome number was between 68 and 70 in all the species. In several metaphases, however, the chromosome number was much lower than expected due to chromosome loss. The largest counts were considered for establishing the chromosome number.

The preparations were of good quality and the karyotypes of some species are shown in Figures 1-8.

Table I shows the chromosome arms ratio and morphology data for the different species. Results for *A. autumnalis* and *A. pretrei* are not shown because the preparations were not of a sufficiently high quality to allow a detailed analysis. They followed, however, the pattern presented by the *Amazona* genus.

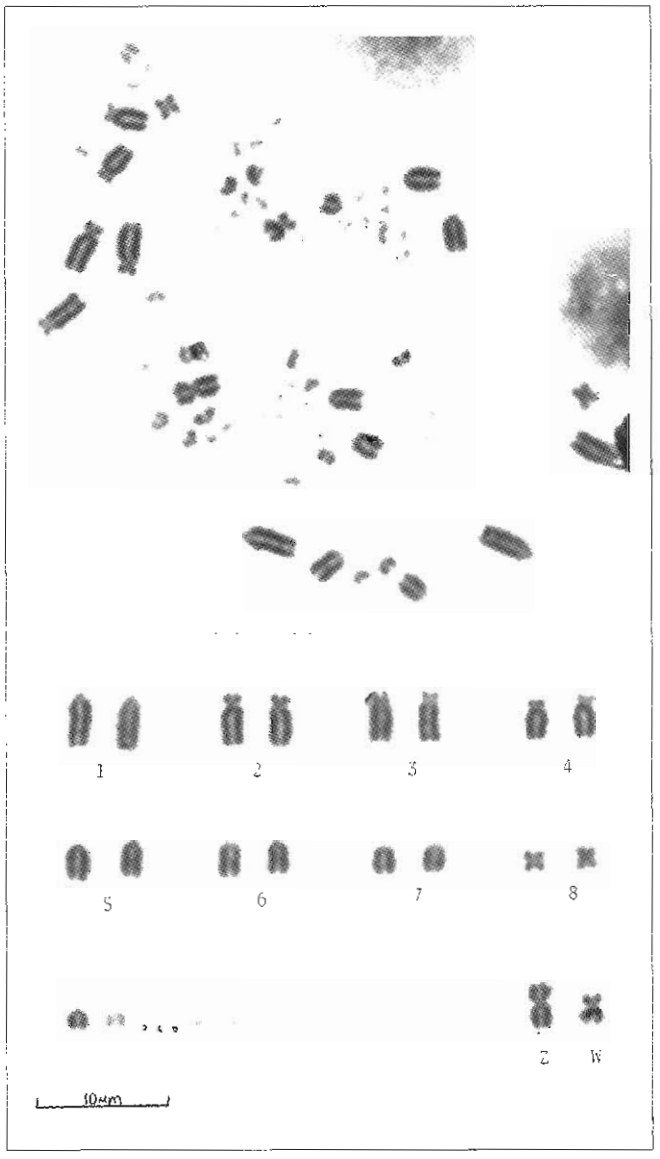

Figure 1 - Karyotype of a female *Amazona brasiliensis*.

DISCUSSION

A great similarity was observed among 11 of the 12 species studied, including *A. autumnalis* and *A. pretrei* which were only visually analyzed. In these species chromosome pairs one, five, six and seven were telocentric. Chromosome two was submetacentric except in *A. amazonica* and *A. festiva* where it was subtelocentric. Pair three was subtelocentric in *A. aestiva*, *A. amazonica*, *A. brasiliensis*, *A. kawalli* and *A. rhodocorytha* and submetacentric in *A. festiva*, *A.*

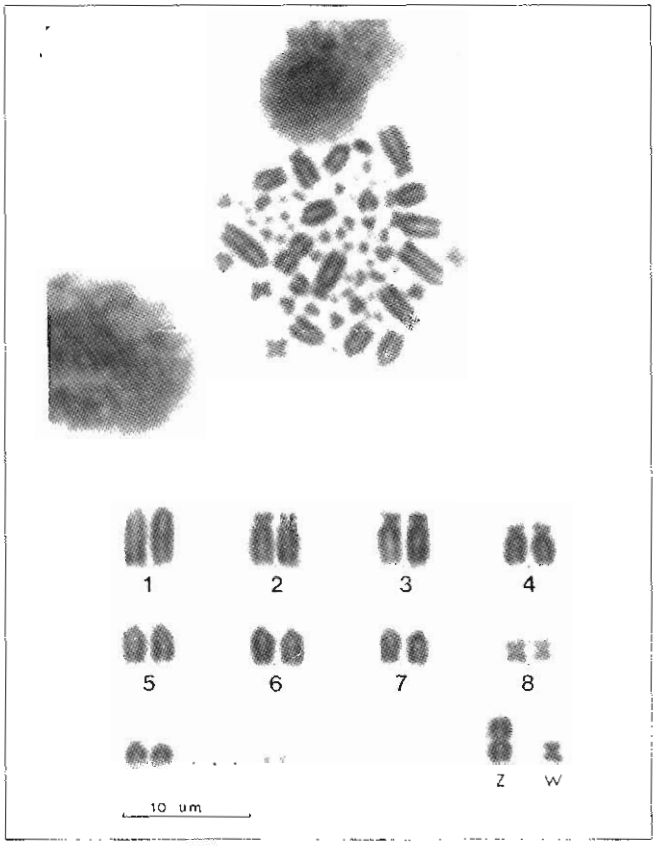

Figure 2 - Karyotype of a female *Amazona farinosa*.

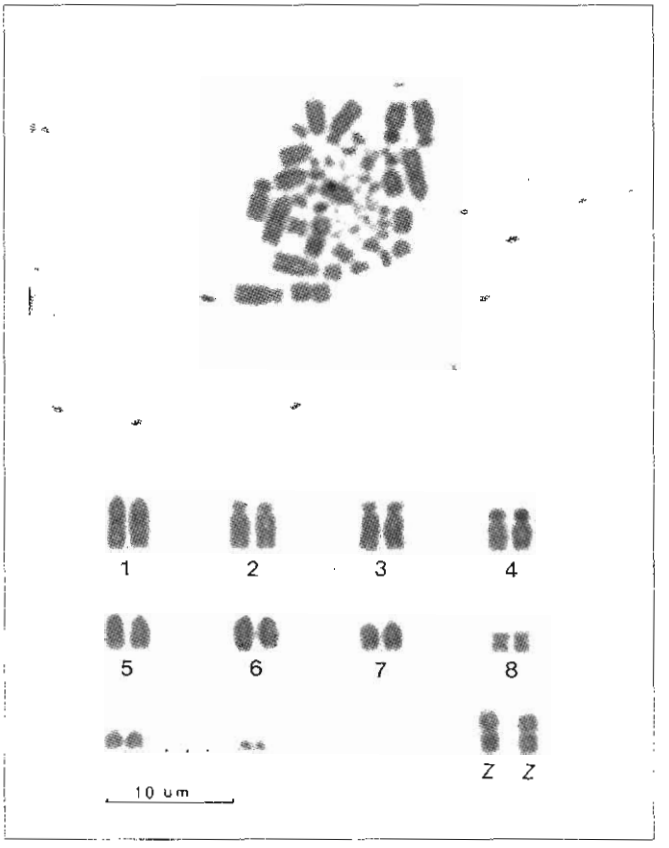

Figure 3 - Karyotype of a male *Amazona festiva*.

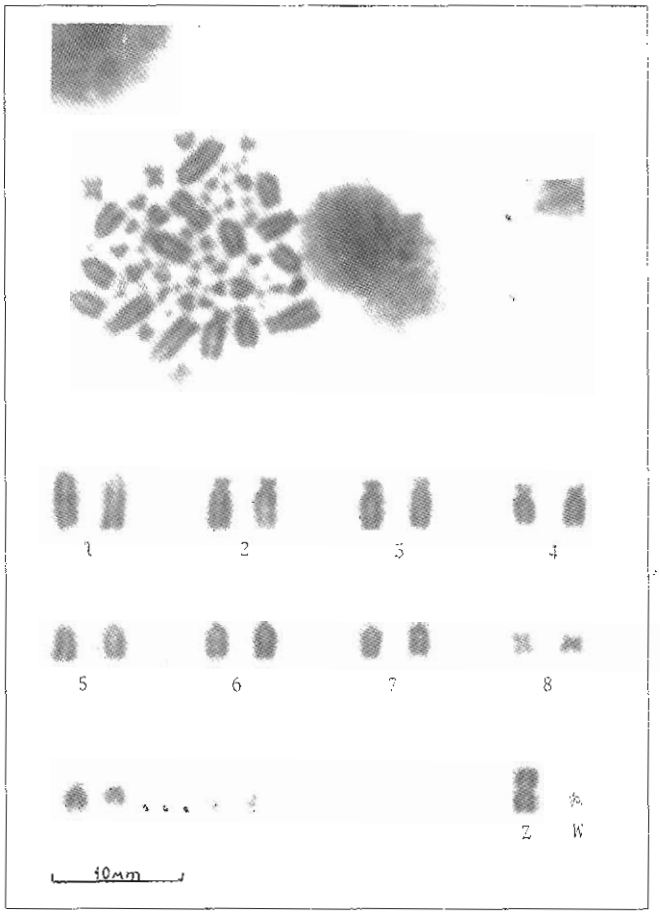

Figure 4 - Karyotype of a female *Amazona kawalli*.

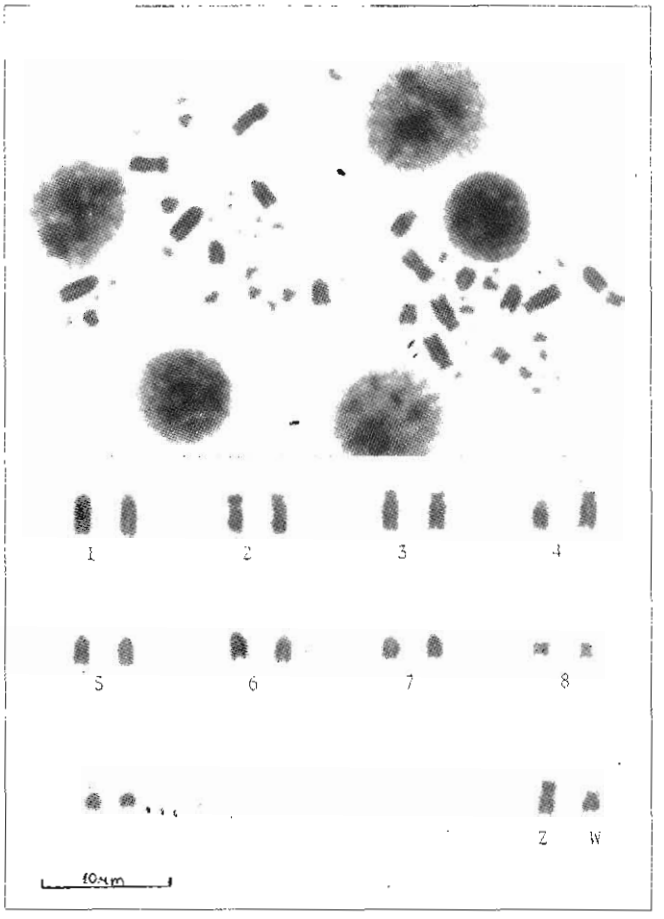

Figure 5 - Karyotype of a female *Amazona rhodocorytha*.

Figure 6 - Karyotype of a female *Amazona vinacea*.

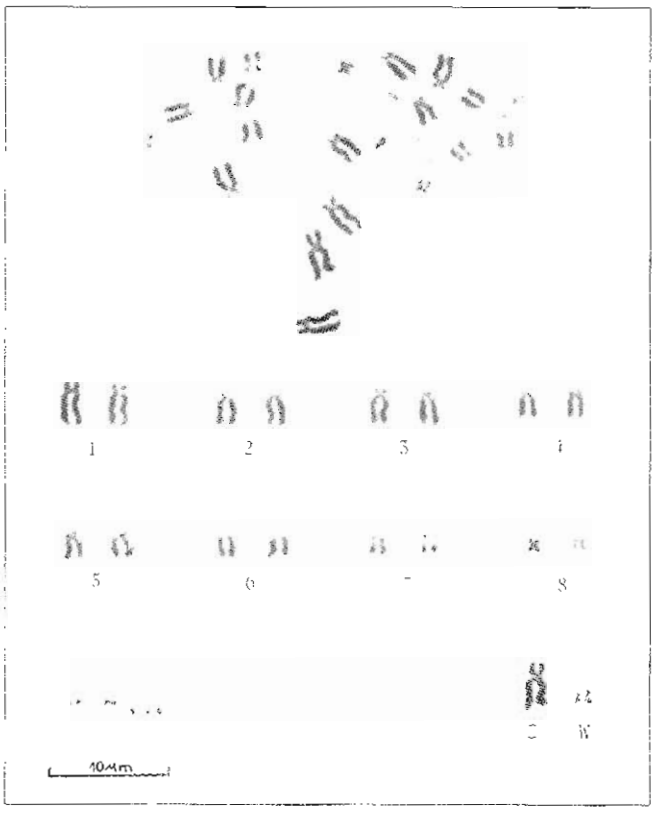

Figure 7 - Karyotype of a female *Salvatoria xanthops*.

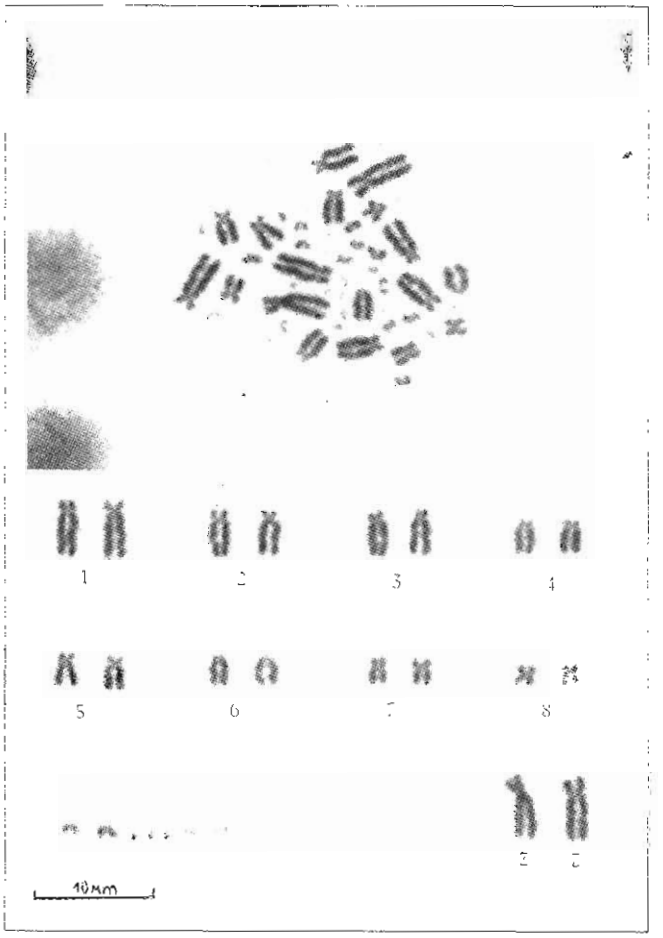

Figure 8 - Karyotype of a male *Salvatoria xanthops*.

ochrocephala, *A. farinosa* and *A. vinacea*. Chromosomes four and eight were submetacentric and metacentric, respectively, in all the species. The sex chromosome Z and W were metacentric in all the species and ranked approximately fourth and eighth in pair size, respectively.

These data confirm, in part, early findings reported in the literature. Aquino and Ferrari (1990) indicated pairs 2, 3, 4 and 5 as subtelocentric for *A. aestiva* and *A. amazonica*. In the present study the second pair was submetacentric in the majority of the species and the fourth pair was submetacentric in all species. The fifth pair was clearly telocentric. These authors did not quote the method used to classify the chromosomes, which hampers data comparison. Schmutz and Prus (1987) classified chromosomes 3, 4 and 5 as metacentric in *A. aestiva*. Boer and Belterman (1981) classified the chromosomes 2, 3 and 4 in *A. ochrocephala* as subtelocentric with arms ratios of 5.0,

Table I - Morphology, mean and range of variation of the arms ratio of the macrochromosomes of the different species of the *Amazona* genus. (T = telocentric; ST = subtelocentric; SM = submetacentric and M = metacentric).

Species	1	2	3	4	5	6	7	8	Z	W
Amazona aestiva	T-oo	SM-2.97 (2.50-4.08)	ST-3.36 (2.50-4.31)	SM-2.87 (2.29-3.60)	T-oo	T-oo	T-oo	M-1.14 (1.00-1.36)	M-1.19 (1.03-1.44)	M-1.20 (1.04-1.35)
Amazona amazonica	T-oo	ST-3.41 (2.88-4.08)	ST-3.42 (3.00-3.93)	SM-2.57 (2.27-3.00)	T-oo	T-oo	T-oo	M-1.09 (1.00-1.31)	M-1.31 (1.11-1.75)	M-1.48 (1.20-1.85)
Amazona brasiliensis	T-oo	SM-2.79 (2.26-3.61)	ST-3.42 (2.50-4.17)	SM-2.96 (2.46-4.00)	T-oo	T-oo	T-oo	M-1.19 (1.04-1.37)	M-1.21 (1.04-1.41)	M-1.32 (1.17-1.45)
Amazona festiva	T-oo	ST-3.09 (2.33-3.68)	SM-2.61 (2.14-3.09)	SM-2.07 (1.47-2.81)	T-oo	T-oo	T-oo	M-1.16 (1.00-1.28)	M-1.07 (1.02-1.14)	-
Amazona kawalli	T-oo	SM-2.78 (2.66-2.89)	ST-3.11 (2.91-3.30)	SM-2.44 (2.40-2.47)	T-oo	T-oo	T-oo	M-1.02 (1.00-1.03)	M-1.00	M-1.11
Amazona ochrocephala	T-oo	ST-3.24 (2.03-5.32)	SM-2.95 (2.44-3.41)	SM-2.84 (2.00-3.57)	T-oo	T-oo	T-oo	M-1.19 (1.09-1.28)	M-1.17 (1.07-1.27)	-
Amazona rhodocorytha	T-oo	SM-2.77 (2.00-3.19)	ST-3.12 (2.79-3.40)	SM-2.45 (1.77-3.19)	T-oo	T-oo	T-oo	M-1.22 (1.00-1.55)	M-1.23 (1.11-1.43)	M-1.48 (1.23-1.65)
Amazona farinosa	T-oo	SM-2.50 (2.45-2.54)	SM-2.99 (2.88-3.09)	SM-2.10 (1.89-2.30)	T-oo	T-oo	T-oo	M-1.07 (1.00-1.13)	M-1.07 (1.04-1.09)	
Amazona vinacea	T-oo	SM-2.54 (2.03-2.88)	SM-2.55 (1.90-3.13)	SM-2.28 (1.67-2.93)	T-oo	T-oo	T-oo	M-1.04 (1.00-1.08)	M-1.46 (1.28-1.64)	M-1.04 (1.00-1.08)
Salvatoria xanthops	ST-3.11 (2.24-4.47)	SM-2.71 (1.46-3.71)	SM-2.80 (1.97-3.56)	SM-2.52 (1.74-3.31)	SM-2.25 (1.30-2.84)	T-oo	M-1.37 (1.14-1.68)	M-1.18 (1.00-1.88)	SM-2.44 1.59-3.74)	SM-1.98 (1.81-2.13)

5.0 and 4.5, respectively. These data are very different from those presented here (3.24, 2.95 and 2.84). Only one or two metaphases were analyzed for some species, in these reports.

The *A. xanthops* karyotype differed considerably from the general pattern of the *Amazona* genus. Chromosomes 1, 5 and 7 are subtelocentric, submetacentric and metacentric, respectively, while in other *amazona* species they are telocentric. The Z chromosome in this species is submetacentric and the largest in the karyotype. This morphology is unique in the Psittacidae analyzed up to now. The W chromosome was submetacentric and approximately the sixth pair in size, which is also unique.

Sick (1984) reported in the morphological analysis of *A. xanthops* that its "inclusion of the species in the *Amazona* genus may not be correct". Based on his data and the results presented the species *A. xanthops* was removed from the *Amazona* genus.

This decision can be cytogenetically justified by the chromosomic conservation found in the *Amazona* genus. Changes at the level of those which were found in *A. xanthops* cannot be explained except by a much earlier separation in the evolutionary tree of the Psittacidae family.

The confirmation of the genus *Salvatoria* created by Ribeiro (1920) is recommended from the data shown, naming the species *Salvatoria xanthops*. The inclusion of the species *S. xanthops* in a monospecific genus is necessary because of its great karyotypic distance from the other Psittacidae.

The genus which is closest to the *Salvatoria* is *Pionus* (Lucca *et al.*, 1991) in spite of the differences in the chromosomes 7, 8, Z and W.

ACKNOWLEDGMENTS

The authors thank the animal keepers Maurício G.F. dos Santos, Nelson M. Kawall, Luis Maluf and Roberto Martins and the Zoos at Bauru, Sorocaba, Americana, Piracicaba and Rio de Janeiro for the supply of biological material for this study. We are also grateful to the technicians João Airton Boer and Paulo Antonio Tosta for help in the laboratory and to Prof. Jacques Vielliard for help in the taxonomic review of *Salvatoria xanthops*.

Publication supported by FAPESP.

RESUMO

Os cariótipos de 12 espécies de Psittacidae do gênero *Amazona* foram estudados: *A. aestiva, A. amazonica, A. brasiliensis, A. autumnalis, A. farinosa, A. festiva, A. kawalli, A. ochrocephala, A. pretrei, A. rhodocorytha, A. vinacea* e *A. xanthops*. Foi utilizada a cultura de curta duração de polpa de pena, para obtenção das metáfases. Onze das 12 espécies analisadas foram homogêneas cariotipicamente, com apenas algumas divergências nos cromossomos 2 e 3. A espécie *A. xanthops* apresentou importantes diferenças cariotípicas em relação ao gênero *Amazona*. Desta forma foi revalidado o gênero *Salvatoria* (Ribeiro, *Rev. Mus. Paul. 12*: 1-82, 1920), permanecendo *Salvatoria xanthops*.

O trabalho evidenciou o conservadorismo cromossômico no gênero *Amazona* e a necessidade de maiores esforços no estudo dos Psittacidae, com vistas a citotaxonomia.

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(Received August 18, 1994)