Genetics and Molecular Biology, 27, 4, 522-528 (2004)

www.sbg.org.br

Research Article

Chromosomal similarity between the Scaly-headed parrot (*Pionus maximiliani*), the Short-tailed parrot (*Graydidascalus brachyurus*) and the Yellow-faced parrot (*Salvatoria xanthops*) (Psittaciformes: Aves): A cytotaxonomic analysis

Renato Caparroz[1] and José Maurício Barbanti Duarte[2]

[1]*Instituto Pau Brasil de Historia Natural, Arujá, SP, Brazil.*
[2]*Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias de Jaboticabal, Departamento de Zootecnia, Jaboticabal, SP, Brazil.*

Abstract

Behavior, morphology, allozyme studies and DNA hybridization and sequencing data all suggest the independent evolution of the Old and New World parrots and support tribe status for the American species, although the phylogenetic relationships within this tribe are still poorly understood. A previous study has shown that the Yellow-faced parrot (*Amazona xanthops* Spix, 1824) exhibits large karyotypic differences compared to the other *Amazona* species and suggested that this species should be renamed *Salvatoria xanthops*, although the relationships between *S. xanthops* and the other New World parrots remain unclear. In the present work, we describe the karyotype of the Scaly-headed parrot (*Pionus maximiliani* Kuhl, 1820) and the karyotype and C-banding pattern of the Short-tailed parrot (*Graydidascalus brachyurus* Kuhl, 1820) and compare them to the karyotype and C-banding pattern of *S. xanthops*, as well as to the karyotypes of other New World parrots. The chromosomal similarity between these three species and the karyotypic differences between them and other New World parrots suggest that *G. brachyurus* and *S. xanthops* are sister species and are most closely related to members of the genus *Pionus*.

Key words: chromosome, karyotype, cytotaxonomy, psittaciformes, parrots.

Received: May 7, 2003; Accepted: March 5, 2004.

Introduction

New World parrots not only share many unique characters (e.g. chicks with imperforated ear canals and their one-legged copulatory stance) but are also wide biochemically distant from other parrots (Mainardi, 1962; Gysels, 1964), leading Smith (1975) to suggest that the New World parrots appear to form a monophyletic radiation. Allozyme (Randi and Bertagnolio, 1990) and DNA hybridization (Sibley and Alquist, 1990) studies also support the separation of Old and New World parrots, and Collar (1997) has suggested that the approximately 150 species of New world parrots should be accorded the status of a tribe (Arini). The separation of Old and New World parrots is also supported by DNA sequencing studies (Miyaki *et al*., 1998), although the exact phylogenetic relationships within this group are still poorly understood.

Send correspondence to Renato Caparroz. Instituto Pau Brasil de Historia Natural, Rua Nhu Guaçú 71, Nova Pirajussara, 04625-000 São Paulo, SP, Brazil. E-mail: renatocz@yahoo.com.br.

Chromosomal analyses have produced data that could help to shed light on the evolutionary and taxonomic relationships within the New World parrots. Generally, where structural differences in karyotypes exist between various species of a group it is possible to form hypotheses as to their phylogenetic relationships (Van Dongen and De Boer, 1984). The American parrots is one of the most cytogenetically studied groups of New World birds (Lucca, 1974, 1984, 1985; De Boer and Belterman, 1980; Van Dongen and De Boer, 1984; Schmutz and Prus, 1987; Valentine, 1987; Aquino and Ferrari, 1985, 1990; Duarte and Giannoni, 1990; Lucca *et al*., 1991; Archangelo *et al*., 1995; Duarte and Caparroz, 1995; Rocha *et al*., 1995; Goldschmidt *et al*., 1997; Francisco *et al*., 2001; Lunardi *et al*., 2003), the species so far analyzed exhibiting a heterogeneous karyotypic morphology among some genera in contrast with several other avian groups in which very uniform karyotypes are found.

Duarte and Caparroz (1995) showed that the Yellow-faced parrot (*Amazona xanthops* Spix, 1824), a species

endemic to eastern and central Brazil (Forshaw, 1989), exhibited large karyotypic differences not only with the other species of the genus *Amazona* but also all the other New World parrots that have so far been described, while the morphological analysis conducted by Sick (1984) led him to state that the `inclusion of (*A. xanthops*) in the *Amazona* genus may not be correct. Duarte and Caparroz (1995) used these differences to suggest that *A. xanthops* Spix, 1824 should be excluded from the genus *Amazona* and renamed *Salvatoria xanthops* (as first used by Ribeiro, 1920) to form the monotypic genus *Salvatoria* which is cytogenetically most closely related to the genus *Pionus.* Despite these findings, a clear understanding of the relationships between *S. xanthops* and the other New World parrots remains unclear.

In this paper we describe the karyotype of the Scaly-headed parrot (*Pionus maximiliani* Kuhl, 1820), which occurs from northern Argentina to the northern Brazilian states of Ceará, Piauí and Goiás (Forshaw, 1989), and the karyotype and C-banding pattern of the Short-tailed parrot (*Graydidascalus brachyurus* Kuhl, 1820) which inhabits the forests of Colombia, Ecuador, Peru and the Brazilian Amazon basin (Forshaw, 1989). We also discuss the phylogenetic relationships of *P. Maximiliani* and *G. brachyurus* in relation to the previously reported karyotype of *S. xanthops* (Duarte and Caparroz, 1995) and other New World parrots.

Material and Methods

We investigated four male and three female *Graydidascalus brachyurus* Kuhl, 1820, one male and two female *Pionus maximiliani* Kuhl, 1820 and two female *S. xanthops* captive parrots maintained in Zoos (see acknowledgments for details).

Metaphase chromosomes were obtained using the short-term feather-pulp culture technique as described by Duarte and Caparroz (1995). Briefly, tissue extracted from the bulbs of young feathers was cultured for six hours at 39 °C in a complete culture medium containing 8.0 mL of Mc Coy's medium, 1.0 mL of inactivated horse serum and 0.6 mL of phytohemaglutinin to which 0.0016% of colchicine was added for the last two hours. After culture, the cells were exposed to hypotonic KCl solution (0.075 M) for 15 min at 39 °C and then fixed in 3:1 methanol/acetic acid and the cell suspension dropped onto clean slides and stained with 3.5% Giemsa-phosphate (pH 6.8).

The C-banding patterns were obtained for the *G. brachyurus* and *S. xanthops* specimens (the chromosomes of the *P. maximiliani* specimens were not C-banded) using a modification of the barium hydroxide ($Ba(OH)_2$) method (Sumner, 1972) in which slides containing cells with metaphase chromosomes were placed in 0.01 M aqueous HCl for 20 min at room temperature and then into 5% aqueous barium hydroxide for 7 to 15 min at 37 °C followed by washing with 0.01 M aqueous HCl for 30 s to remove excess barium hydroxide. After washing, the slides were placed into 2 x SSC (salina sodium-citrate solution) for one hour at 57 °C and then stained with 3.5% Giemsa-phosphate (pH 6.8).

Chromosome biometry was used to determine the arm-ratio (AR) of the chromosomes which were then classification according to centromeric position using the nomenclature of Levan *et al*. (1964).

Results

The metaphases obtained were of good quality and the karyotypes of all the parrots sampled could be established. The diploid number was 72 for *P. maximiliani* and 64 for *G. brachyurus*, although several metaphases had a lower than expected chromosome number due to chromosomes loss and in this case the highest chromosome counts were used to establish the diploid number.

In *P. Maximiliani* the first seven chromosome pairs were macrochromosomes and the remaining pairs microchromosomes (Figure 1), with the first five autosomes being submetacentric (AR = 2.21, 2.39, 1,87, 1,75 and 1,72) and the sixth pair telocentric. The microchromosomes of *P. maximiliani* were telocentric up to the point to which their morphology could be identified, while the Z-chromosome was submetacentric (AR = 1.87) and the largest in the karyotype, and the W-chromosome was submetacentric (AR = 1.69) and equivalent in size to the autosomic pair 4.

The karyotype of *G. brachyurus* comprised 18 macro and 46 microchromosomes (Figure 2), with the first five autosomes being submetacentric (AR = 2.47, 2.71, 2.07, 2.17 and 1.74) and the sixth pair telocentric while pairs 7 (AR = 1.25) and 8 (AR = 1.14) were metacentric. The *G. brachyurus* Z-chromosome was submetacentric (AR = 2.63) and the largest in the karyotype, while the W-chromosome was submetacentric (AR = 2.26) and equivalent in size to the autosomic pair 4. Except for the presence of two small pairs (7 and 8) of metacentric macrochromosomes the chromosome morphology of *G. brachyurus* was very similar to that of *P. maximiliani*, and both species showing a very sharp boundary between macro and microchromosomes.

The C-banding patterns of *G. brachyurus* and *S. xanthops* revealed centromeric bands in almost all chromosomes (Figure 3). However, the C-bands were more pronounced in *S. xanthops* than in *G. brachyurus*. In both species, the W-chromosome presented a conspicuous pattern with the short-arm and the pericentromeric region of the long arm being entirely heterochromatic.

Discussion

Within the order Psittaciformes, 69 species belonging to 36 genera from the families Cacatuidae (9 species) and Psittacidae (60 species) have been studied cytologically. Forty-one species of Psittacidae are from the New World

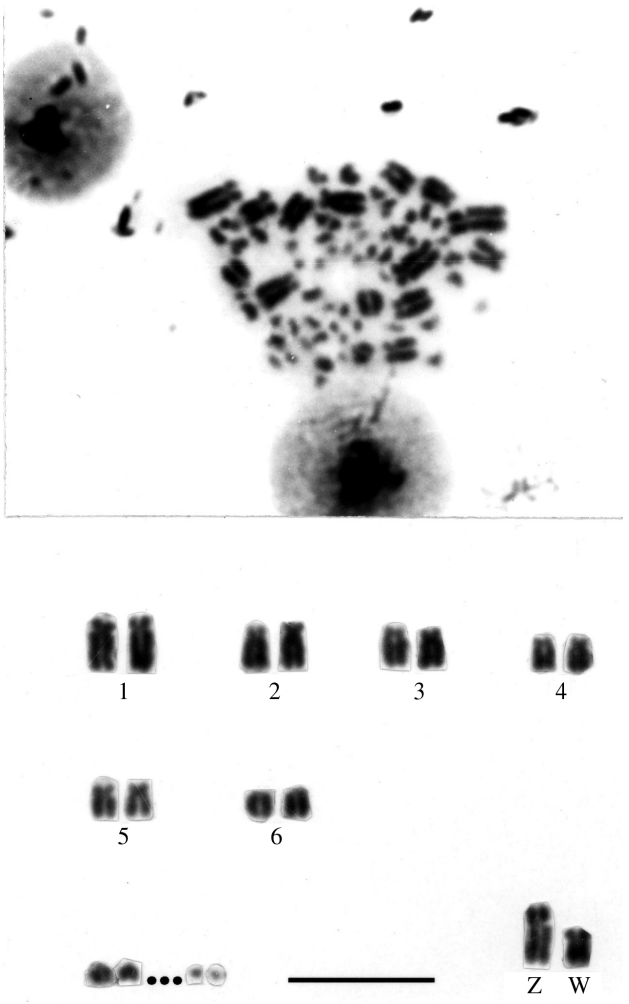

Figure 1 - Conventionally stained mitotic metaphase and female karyotype showing the macrochromosomes and the first and last pairs of microchromosomes of *Pionus maximiliani*. Bar = 10 µm.

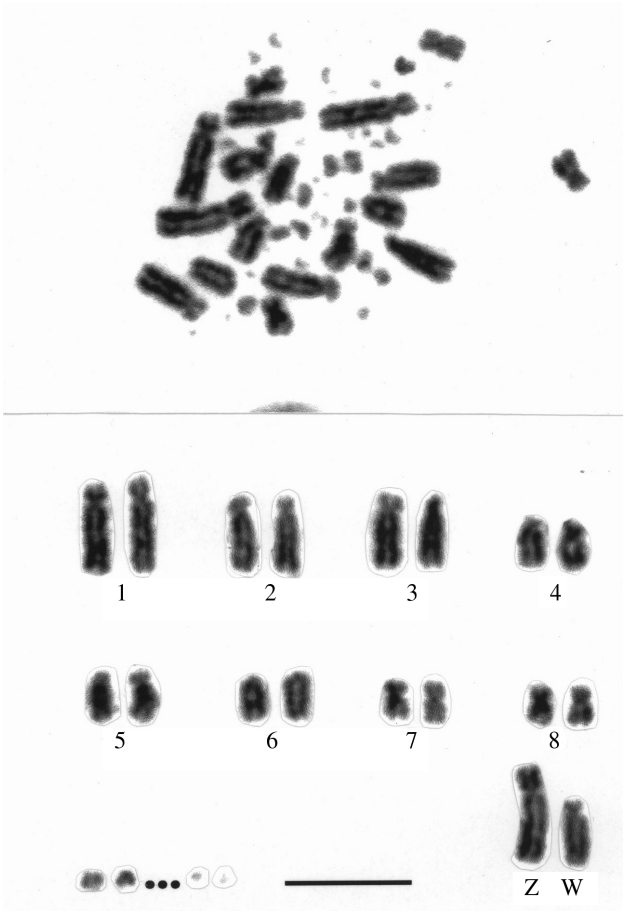

Figure 2 - Conventionally stained mitotic metaphase and female karyotype showing the macrochromosomes and the first and last pairs of microchromosomes of *Graydidascalus brachyurus*. Bar = 10 µm.

and cytological studies have revealed at least three main karyotypic patterns in the New world parrots.

One karyotype pattern is predominant in the genera *Anodorhynchus* (Lunardi *et al*., 2003), *Ara* (Van Dongen and De Boer, 1984; Francisco and Galetti-Jr, 2001), *Cyanopsitta* (Duarte and Giannoni, 1990), *Aratinga* (Lucca, 1984; Lucca *et al*., 1991; Goldschmidt *et al*., 1997), *Guaruba* (Goldschmidt *et al*., 1997), *Nandayus* (Francisco and Galetti-Jr, 2001), *Propyrrhura* (Francisco and Galetti-Jr, 2001), *Pionites* (Francisco *et al*., 2001) and *Deroptyus* (Lunardi *et al*., 2003). This karyogram is characterized by the presence of predominantly biarmed macrochromosomes consisting of about 10 autosomic pairs of which pair 1 is a metacentric conserved both in terms of morphology and size, pairs 2, 3, 4 and 5 vary from submetacentric to subtelocentric, pair 6 from submetacentric to telocentric, pairs 7 and 8 from metacentric to submetacentric and pairs 9 and 10 metacentric to telocentric.

Another karyotype pattern occurs in the species of the genus *Amazona*, and is principally characterized the presence of 8 autosomic macrochromosome pairs with a significant number of telocentric macrochromosomes, including pair 1. Pairs 2, 3 and 4 vary from submetacentric to subtelocentric, and pair 8 is metacentric (De Boer and Belterman, 1980; Van Dongen and De Boer, 1984; Schmutz and Prus, 1987; Aquino and Ferrari, 1990; Lucca *et al.,* 1991; Duarte and Caparroz, 1995).

A further karyotype pattern is observed in the species of the genus *Pionus* (Lucca *et al*., 1991) and also in *Salvatoria xanthops* (Duarte and Caparroz, 1995), and consists between 6 and 8 pairs of macrochromosomes, of which pairs 1, 2, 3 and 4 are submetacentric or subtelocentric. Chromosome pair 5 was described as being subtelocentric in *Pionus maximiliani*, *P. menstruus* and *P. senilis* while in *P. seniloides* it was described as telocentric (Lucca *et al*., 1991). However, pairs 5 and 6 are very similar in size in all *Pionus* species so far studied and it transpires that the *P. seniloides* pair 5 was erroneously identified as pair 6 (and pair 6 as pair 5), which means that the *P. seniloides* pair 5 is really subtelocentric and pair 6 telo-

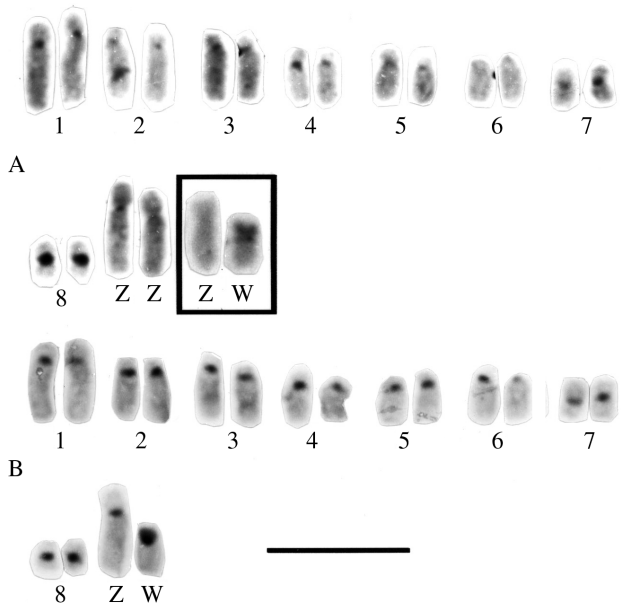

Figure 3 - C-banding patterns of the macrochromosomes of (A) a *Graydidascalus brachyurus* male and (B) a *Salvatoria xanthops* female. A *G. brachyurus* ZW female sex chromosomes is shown in the insert. Bar = 10 µm.

centric, this being true for all *Pionus* species so far studied as well as for *S. xanthops*. In addition, Duarte and Caparroz (1995) report the presence of two small metacentric pairs (pairs 7 and 8) in *S. xanthops* that have been found to be absent in all species of the genus *Pionus* so far karyotyped.

The *P. maximiliani* karyotype described here is different from the karyotype previously reported for this species and is also different from *P. menstruus*, *P. senilis* and *P. seniloides* (Lucca *et al*., 1991). In all *Pionus* so far karyotyped, the first four autosome and the Z-chromosome pairs are subtelocentric while in the *P. maximiliani* specimens analyzed by us they were submetacentric. The karyotypic differences observed between *P. maximiliani* (previously described), *P. menstruus*, *P. senilis* and *P. seniloides* and *P. maximiliani* as described by us in this paper could be due to differences in how the arm-ratio was calculated method leading to divergent classification of the same chromosome pair, because of which we consider that these morphological differences are not significant. In addition, the Z-chromosome of *P. maximiliani*, *P. menstruus* and *P. senilis* was reported by Lucca *et al*. (1991) as being equivalent in size to pair 4, although since only one male of each species was analyzed by Lucca *et al*. (1991) it appears possible that the largest chromosome pair of these *Pionus* species correspond to the Z-chromosomes as observed by us in *P. maximiliani.*

On the other hand, the sexual chromosomes of *P. seniloides* are very different from those observed by Lucca *et al*. (1991) in other *Pionus* and by us in *P. maximiliani*. Since Lucca *et al.* (1991) analyzed one female *P. seniloides* they were able to identify the sexual chromosome pair in this species and found that the Z-chromosome of *P. seniloides* was subtelocentric and of the same size as pair 4, while the W-chromosome was metacentric and smaller than pair 6. The differences in the sex chromosomes between these species appears to be very significant for understanding the phylogenetic relationships within the genus *Pionus* (to be discussed below).

The *G. brachyurus* karyotype described for the first time in this paper is very similar to that previously reported for *S. xanthops* (Duarte and Caparroz, 1995), except for the diploid number, which is 64 in *G. brachyurus* and 68 in *S. xanthops*, and the largest autosome pair which was described as subtelocentric in *S. xanthops* (Figure 4). This karyotypic similarity between *G. brachyurus* and *S. xanthops* suggests that these two species are likely to be closely related, and the similarity between the C-banding patterns (especially for the W-chromosome) observed in these two species also supports this similarity. Both the short arm and the pericentromeric region of the long arm of the W-chromosome are constitutively heterochromatic in *G. brachyurus* and *S. xanthops* while in all other New World parrots studied the W-chromosome is largely C-positive. Very little is known about the patterns of heterochromatin distribution among the Psittaciformes because C-banding information is only available for few species (Mengden, 1981; Lucca, 1983, 1984; Lucca and De Marco, 1983; Aquino, 1987; Aquino and Ferrari, 1990; Christidis *et al*., 1991), all of which are South American. However, the data available indicate that changes in both the quantity and the distribution of heterochromatin have played a significant role in the chromosomal evolution of this order (Christidis *et al*., 1991). The close relationship between *S. xanthops* and *G. brachyurus* is also consistent with the morphological similarity between these species reported previously (Ribeiro, 1920). Furthermore, according to a recent molecular phylogenetic study involving New World parrots (Russello and Amato, 2003) the genus *Amazona* is not a monophyletic group, with *S. xanthops* forming a well-supported sister group relationship with *G. brachyurus* to the exclusion of all other Amazon parrots.

The karyotypes of *G. brachyurus* and *S. xanthops* are more similar to that described here for *P. maximiliani* and to those reported by Lucca *et al*. (1991) for other *Pionus* species than to those reported for other New World parrots. The two metacentric macrochromosomes of *G. brachyurus* and *S. xanthops* (pairs 7 and 8), which are absent in all *Pionus* species, could be a result of a reduction in the diploid number (72 in all *Pionus* species but 64 in *G. brachyurus* and 68 in *S. xanthops*) by Robertsonian translocations between larger microchromosomes. Van Donger and De Boer (1984) pointed out that the small metacentric pairs observed in *Melopsittacus undulatus* and *Ara* species were likely to have been formed by Robertsonian translocation between larger microchromosomes, while Christidis *et al*. (1991) concluded that

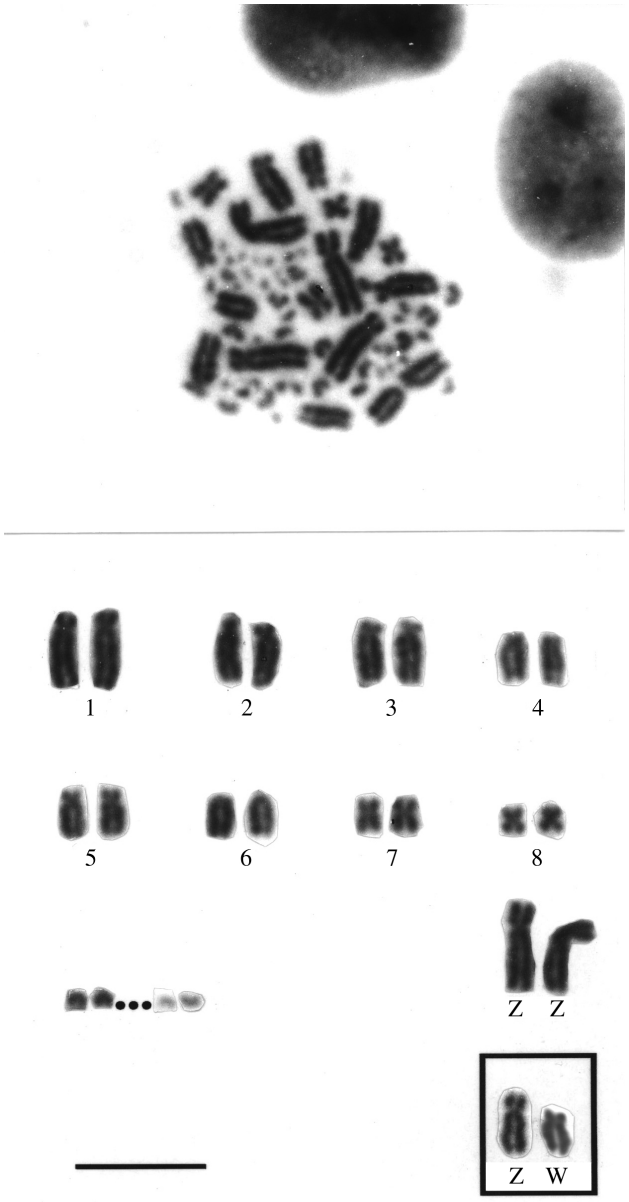

Figure 4 - Conventionally stained mitotic metaphase and male karyotype showing the macrochromosomes and the first and last pairs of microchromosomes of *Salvatoria xanthops*, in conventional staining. In the insert, the ZW female sex chromosomes. Bar = 10 µm.

a karyotype lacking biarmed autosomes is ancestral for the Cacatuidae and that the derived karyotypes were evolved by micro-macrochromosome fusions.

The characteristics of the sexual chromosomes of *G. brachyurus* and *S. xanthops,* which are shared by *Pionus* (except by *P. seniloides*), are also unique. In all these species, the Z-chromosome is submetacentric and is the largest chromosome in the karyotype while the W-chromosome is submetacentric and equivalent in size to macrochromosome pair 4. The Z-chromosome appears to be conserved in all the remaining New World parrots as it is metacentric and similar in size, corresponding to chromosome pair 4 or 5. The W-chromosome is morphologically more variable among the New World parrots than the Z-chromosome, the W-chromosome having been identified as metacentric in most of the New World parrots, submetacentric in *Anodorhynchus hyacinthinus* (Lunardi *et al*., 2003), subtelocentric in *Aratinga leucophthalmus* (Lucca, 1984) and telocentric in *Propyrrhura maracana, Aratinga auricapilla* and *Deroptyus accipitrinus* (Francisco and Galetti, 2001; Lunardi *et al*., 2003). However, the W-chromosome showed a size similar to the smallest macro-autosomes in all the species studied.

Based on the chromosomal similarity between *G. brachyurus*, *S. xanthops* and some species of the genus *Pionus*, especially for the sexual chromosomes (except for *P. seniloides*), and the karyotypic differences from the other New World parrots, it appears likely that these species are closely related. In addition, the fact that at least for the New World parrot analyzed so far and belonging to the same genus, such as *Amazona*, *Ara* and *Aratinga*, have very similar karyotypes supports this point of view. Since *G. brachyurus* and *S. xanthops* probably share two derived small metacentrics, it seems likely that chromosome rearrangements occurred after divergence from the common ancestor of *G. brachyurus* and *S. xanthops* and members of the genus *Pionus*. This being the case, *G. brachyurus* and *S. xanthops* are considered by us to be sister species closely related to the genus *Pionus*.

As mentioned above, the sexual chromosomes of *P. seniloides* are different in morphology and size from those described for the other *Pionus* species and seem more similar to those reported for the New World parrots so far studied and also to some Australian parrots (Van Dongen and De Boer, 1984; Christidis *et al*., 1991). Based on these data, it seems possible that the chromosome rearrangements responsible for the formation of the karyotypic pattern shared by *P. maximiliani*, *P. menstruus*, *P. senilis*, *G. brachyurus* and *S. xanthops* may have occurred after the divergence between the common ancestor of these five latter species and *P. seniloides*. If this is indeed the case, the genus *Pionus*, as currently described, is not monophyletic, although the possibility of karyotypic convergence in *P. seniloides,* while unlikely, cannot be discard.

A close phylogenetic relationship between *S. xanthops*, *G. brachyurus* and some *Pionus* species (*P. menstruus* and *P. senilis*) has also been observed in some molecular analyses (Miyaki *et al*., 1998; Russello and Amato, 2003). However, as the taxonomic sampling of species in these studies was specifically tailored to address the question of monophyly of genera or groups determined *a priori*, these studies were unable to investigate the species level relationships of the genus *Pionus*. A forthcoming phylogenetic analysis of all *Pionus* species is expected to add to our understanding of the phylogenetic relationships within this genus.

Finally, to confirm the results presented here, detailed analyses of inter-specific homologies based on chromosome banding methods are needed, both in the species already studied and in many other species not investigated in our study. The present study, and others cited above, demonstrate the usefulness of using chromosomal data to reconstruct evolutionary relationships in the Psittaciformes. Moreover, investigations involving more sophisticated chromosome staining methods, as well as employing other methodologies, such as DNA sequencing, will likely prove useful in fully elucidating the phylogenetic relationships within the Psittaciformes.

Acknowledgments

The authors would like to thank the following Zoos in Brazil for the supply of the biological samples for this study: Zoo Parque (one female *G. brachyurus* sample), the Fundação Rio-Zoo (four males and two females *G. brachyurus* samples), the Zoo of Piracicaba (one female *S. xanthops*) and the Zoo of Sorocaba (one male and two females *P. maximiliani* and one female *S. xanthops* samples) and Celia P. Koiffmann, Cristina Y. Miyaki, Erika Schlenz, Eugene Harris and Lurdes Foresti de Almeida Toledo for helpful discussion. We are also grateful to the technicians João Airton Boer, Paulo Antonio Tosta, Cláudia Irene Emílio de Castro, Waldir Caldeira and Carlos Eduardo Lopes for their help in the laboratory, and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for financial support, and anonymous reviewer commented on the manuscript.

References

Aquino R and Ferrari I (1985) Estudo dos cromossomos de *Ara ararauna* e *Ara macao*. Resumos do XII Congresso Brasileiro de Zoologia, Campinas, Universidade Estadual de Campinas, p 255.

Aquino R and Ferrari I (1990) Chromosome study of *Amazona amazonica* and *A. aestiva* (Aves: Psittaciformes): Determination of chromosome number and identification of sex chromosomes by C-banding methods. Genetica 81:1-3.

Archangelo L, Galetti-Jr PM, Rocha GT and Lucca EJ (1995) Nota sobre a aplicação de fluorocromos AT-e G-C específicos em cromossomos de Araras. Braz J Genet 18:479.

Birt TP, Friesen VL, Green JM, Montevecchi WA and Davidson WS (1992) Cytochrome-*b* sequence variation among parrots. Hereditas 117:62-72.

Christidis L, Shaw DD and Schodde R (1991) Chromosomal evolution in parrots, lorikeets and cockatoos (Aves: Psittaciformes). Hereditas 114:47-56.

Collar NJ (1997) Family Psittacidae (Parrots). In: del Hoyo J, Elliot AE and Sargatal J (eds) Handbook of the Birds of the World. v. 4. Lynx Editions, Barcelona, pp 280-477.

De Boer LEM and Belterman RHR (1980) The somatic chromosomes of three parrots: The Kea (*Nestor notabilis*), the Yellow-headed parrot (*Amazona ochrocephala*) and the Grey parrot (*Psittacus erithacus*). Acta Zool Pathol Antuerphensia 75:9-18.

Duarte JMB and Caparroz R (1995) Cytotaxonomic analysis of Brazilian species of the genus *Amazona* (Psittacidae, Aves) and confirmation of the genus *Salvatoria* (Ribeiro, 1920). Braz J Genet 18:623-628.

Duarte JMB and Giannoni ML (1990) Karyotype of the Little blue macaw *Cyanopsitta spixii* (Psittaciformes: Aves). Rev Bras Genet 13:137-140.

Francisco MR, Lunardi VO and Galetti-Jr PM (2001) Chromosomal evidences of adaptive convergence in the tail morphology of neotropical Psittacidae (Aves: Psittaciformes). Cytologia 66:329-332.

Forshaw JM (1989) The parrots of the world. Lansdowne editions, Sidney, 672 pp.

Goldschmidt B, Nogueira DM, Monsores DW and Souza LM (1997) Chromosome study in two *Aratinga* species (*A. guarouba* and *A. acuticaudata*) (Psittaciformes). Braz J Genet 20:659-662.

Gysels H (1964) A biochemical evidence for the heterogeneity of the family Psittacidae. Bull Soc R Zool Anvers 33:29-41.

Levan A, Fredga K and Sanberg AA (1964) Nomenclature for centromeric position on chromosomes. Hereditas 52:201-220.

Lucca EJ (1974) Cariótipos de 8 espécies de aves. Rev Bras Biol 34:387-392.

Lucca EJ (1983) Constitutive heterochromatin and the structural complexity of chromosomes in Columbiformes and Psittaciformes (Aves). Caryologia 36:373-384.

Lucca EJ (1984) A comparative study of the chromosomes in 5 species of birds from the genus *Aratinga* (Psittaciformes: Aves). Cytologia 49:537-545.

Lucca EJ (1985) The somatic chromosomes of *Brotogeris sanctithomae* and *Brotogeris versicolorus* (Psittaciformes: Aves). Genét Ibér 37:39-45.

Lucca EJ and Marco DA (1983) Chromosomal polymorphism in *Forpus xanthopterygius* (Psittaciformes: Aves). Caryologia 36:355-361.

Lucca EJ, Shirley LR and Lanier C (1991) karyotype studies in twenty-two species of parrots (Psittaciformes: Aves). Rev Bras Genet 14:73-98.

Lunardi VO, Francisco MR, Rocha GT, Goldschmidt B and Galetti-Jr PM (2003) Karyotype description of two Neotropical Psittacidae species: The endangered Hyacinth Macaw, *Anodorhynchus hyacinthinus*, and the Hawk-headed Parrot, *Deroptyus accipitrinus* (Psittaciformes: Aves), and its significance for conservation plans. Genet Mol Biol 26:283-287.

Mainardi D (1962) Studio immunogenetico sulla posizione tassanomica di *Melopsittacus undulatus*. Riv Ital Orn 32:136-140.

Mengden GA (1981) Linear differentiation of the C-band pattern of the W chromosome is snakes and birds. Chromosoma 83:275-287.

Miyaki CY, Matioli SR, Burke T and Wajntal A (1998) Parrot evolution and paleogeographical events: Mitochondrial DNA evidence. Mol Biol Evol 15:544-551.

Randi E and Bertagnolio P (1990) Biochemical relationships in the Psittaciformes. International Ornithological Congress, New Zealand, abstract 686.

Ribeiro AM (1920) Revisão dos psittacídeos brasileiros. Revista do Museu Paulista 12:1-82.

Rocha GT, Santos MS, Amaro RC and Lucca EJ (1995) Análise cromossômica e determinação do sexo de aves ameaçadas de extinção. Braz J Genet 18:479.

Russello MA and Amato G (2003). A molecular phylogeny of Amazona: Implications for Neotropical parrot biogeography, taxonomy, and conservation. Mol Phylog Evol 30:421-437.

Schmutz SM and Prus SE (1987) A cytogenetic study of four species of cockatoos and Amazon parrots. Genetica 74:69-71.

Sibley CG and Alquist JE (1990) Phylogeny and classification of birds - A study in molecular evolution. Yale University Press, New Haven, Conn.

Sick H (1984) Ornitologia Brasileira, Uma Introdução. Editora Universidade de Brasília, Brasília, pp 291-314.

Smith GA (1975) Systematics of parrots. Ibis 117:18-68.

Summer AT (1972) A simple technique for demonstrating centromeric heterochromatin. Expl Cell Res 75:304-306.

Valentine M (1987) Chromosome analysis of maroon bellied and green cheecked conures. AFA Watchbird (June/July):19.

Van Dongen MWM and De Boer LEM (1984) Chromosome studies of 8 species of parrots of the families Cacatuidae and Psittacidae (Aves: Psittaciformes). Genetica 65:109-117.

Associate Editor: Yatiyo Yonenaga-Yassuda