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Chromosomal characteristics and karyotype evolution of Oxyopidae spiders (Araneae, Entelegynae)

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ABSTRACT. We made a cytogenetic analysis of four species of Oxyopidae and compared it with the karyotype data of all species of this family. In *Hamataliwa* sp, the mitotic cells showed 2n♂ = 26+X_1X_2 and telocentric chromosomes. The 2n♂ = 28, which has been described for only one oxyopid spider, is the highest diploid number reported for this family. *Peucetia* species exhibited distinct karyotype characteristics, i.e., 2n♂ = 20+X_1X_2 in *P. flava* and 2n♂ = 20+X in *P. rubrolineata*, revealing interspecific chromosome variability within this genus. However, both *Peucetia* species exhibited telocentric chromosomes. The most unexpected karyotype was encountered in *Oxyopes salticus*, which presented 2n♂ = 10+X in most individuals and a predominance of biarmed chromosomes. Additionally, one male of the sample of *O. salticus* was heterozygous for a centric fusion that originated the first chromosomal pair and exhibited one supernumerary chromosome in some cells. Testicular nuclei of *Hamataliwa* sp and *O. salticus* revealed NORs on autosomal pairs, after silver impregnation. The majority of Oxyopidae spiders have their karyotype differentiated by both reduction in diploid

number chromosome number and change of the sex chromosome system to X type; however, certain species retain the ancestral chromosome constitution 2n = 26+X1X2. The most remarkable karyotype differentiation occurred in *O. salticus* studied here, which showed the lowest diploid number ever observed in Oxyopidae and the second lowest registered for Entelegynae spiders.

Key words: Chromosome fusion; Cytogenetic; Diploid number; Nucleolar organizer region; Sex chromosome system

INTRODUCTION

Spiders are one of the most diverse orders of animals and are certainly the most abundant terrestrial predators (Coddington and Levi, 1991). Cytogenetically, only about 1.5% (Král et al., 2006) of the 41,719 species belonging to the order Araneae (Platnick, 2010) have been analyzed, which showed a wide diversity of diploid chromosome number, ranging from $2n♂ = 7$ to $2n♂ = 96$, and different types of sex chromosome system, e.g., XY, X, X_1X_2, X_1X_2Y, $X_1X_2X_3$, $X_1X_2X_3X_4$ (Araujo, 2007). In species of the monophyletic basal groups, the highest chromosome numbers were verified, such as $2n♂ = 96$ in the suborder Mesothelae and $2n♂ = 86$ in the infraorder Mygalomorphae. In these groups, the karyotypes were composed of biarmed or uniarmed chromosomes (Suzuki, 1954; Srivastava and Shukla, 1986; Řezač et al., 2006). Among the derivative groups included in the infraorder Araneomorphae, the spiders of the Haplogynae lineage presented a predominance of low diploid chromosome numbers that varied between $2n♂ = 7$ and $2n♂ = 37$, a sex chromosome system of the X type, and meta/submetacentric chromosomes. These features contrast with those of the sister-group Entelegynae, which exhibited the highest number of species taxonomically described and cytogenetically investigated (Araujo, 2007; Platnick, 2010). In entelegyne spiders there is a conservation of the diploid number $2n♂ = 42$, a sex chromosome system of the X_1X_2 type, and chromosomes with acro/telocentric morphology (Araujo et al., 2005; Král et al., 2006).

After comparing the karyotype characteristics of the basal and derived species of Araneae, Suzuki (1954) proposed that chromosomal evolution in this order has occurred through reduction in the chromosome number. Later, Rowell (1990) suggested that in spiders there is a peculiar form of karyotype evolution via "all or nothing" fusions. This proposition was based on the fact that intermediate karyotypes that included both acro/telocentric chromosomes and meta/submetacentric chromosomes are rarely encountered in this group. Král et al. (2006) verified that certain related species of entelegyne spiders differ with regard to the diploid number but not in relation to chromosome morphology that is maintained as acro/telocentric. Thus, the authors attributed the reduction in the chromosome number in these spiders to tandem fusions.

Within the Entelegynae lineage, the family Oxyopidae is very interesting because of its predominance of the $2n♂ = 20+X$. This karyotype includes a diploid chromosome number that is relatively low when compared to those of other families of this group and a sex chromosome system that was observed in only 12% of the entelegyne spiders. The chromosome morphology, however, described for 11 species of a total of 21 studied is conserved as acro/telocentric (Table 1). Oxyopidae belongs to the group of spiders known as true lycosoids, whose monophyly is well supported by morphological characters (Silva Davila, 2003). The families that constitute this group form the clade [(Psechridae (Oxyopidae + Senoculidae)) + (Trechaleidae (Lycosidae + Pisauri-

Table 1. Oxyopidae species cytogenetically analyzed.

Species	Chromosomal formula (2n males)	Chromosomal morphology	Collection locality	Reference
Hamataliwa sp	28=26+X_1X_2	26T+X_1X_2T	Brazil	Present study
Nishina generosa Komatsu (nonem nudum)	21=20+X	20A+XA	Japan	Suzuki (1952)
Oxyopes hindostanicus Pocock, 1901	21=20+X	20A+XA	India	Bole-Gowda (1950), Mittal (1961, 1970)
Oxyopes javanus Thorell, 1887	23=22+X	22H+XH	Philippines	Barrion et al. (1989)
Oxyopes lepidus (Blackwall, 1864) under *Oxyopes similaris*	21=20+X	20A+XA	India	Bole-Gowda (1958)
Oxyopes macilentus L. Koch, 1878	21=20+X 23=22+X	20T+XT 22T+XT	Taiwan	Chen (1999)
Oxyopes pandae Tikader, 1969	21=20+X	-	India	Srivastava and Shukla (1986)
Oxyopes ramosus (Martini & Goeze, 1778)	21=20+X	20A+XM	Finland	Hackman (1948)
Oxyopes ratnae Tikader, 1970	21=20+X	20T+XT	India	Datta and Chartterjee (1983, 1989), Parida and Sharma (1987), Sharma and Parida (1987)
Oxyopes rufisternis Pocock, 1901	21=20+X	20A+XA	India	Mittal (1961, 1970)
Oxyopes ryvesi Pocock, 1901	21=20+X	-	India	Sharma et al. (1960)
Oxyopes salticus Hentz, 1845	22=20+X_1X_2	-	-	Painter (1914)
Oxyopes salticus Hentz, 1845	11=10+X 12=11+X/ 13=11+X+B	6M+2SM+2T+XSM 5M+2SM+4T+XSM/ 5M+2SM+4T+XSM+BSM	Brazil	Present study
Oxyopes scalaris Hentz, 1845	21=20+X	-	United States	Tugmon et al. (1990)
Oxyopes sertatus L. Kock, 1877	21=20+X	20A/T+XA/T	Japan, Taiwan	Suzuki (1950, 1952), Igarashi and Kondo (1977), Chen (1999)
Oxyopes shweta Tikader, 1970	21=20+X	-	India	Parida and Sharma (1987), Sharma and Parida (1987)
Oxyopes sushilae Tikader, 1965	21=20+X	-	India	Srivastava and Shukla (1986)
Oxyopes sp	21=20+X	-	India	Sharma et al. (1960)
Oxyopes sp	22=20+X_1X_2	-	India	Mittal (1961)
Oxyopes sp	22=20+X_1X_2	20A+X_1X_2A	India	Mittal (1970)
Oxyopes sp	21=20+X	-	India	Srivastava and Shukla (1986)
Oxyopes sp	21=20+X	-	India	Srivastava and Shukla (1986)
Peucetia flava Keyserling, 1877	22=20+X_1X_2	20T+X_1X_2T	Brazil	Present study
Peucetia rubrolineata Keyserling, 1877	21=20+X	20T+XT	Brazil	Present study
Peucetia viridana (Stoliczka, 1869)	28=26+X_1X_2	26A+X_1X_2A	India	Bole-Gowda (1950), Parida and Sharma (1987), Sharma and Parida (1987)

A = acrocentric; M = metacentric; SM = submetacentric; T = telocentric; H = holocentric; B = supernumerary chromosome.

dae))] (Silva Davila, 2003). In order to gain a better understanding of the chromosomal characteristics and processes of karyotype evolution of Oxyopidae, four species belonging to three different genera were cytogenetically analyzed with standard staining and silver impregnation. It is worth emphasizing that this is the first chromosomal record for oxyopids from Brazilian fauna.

MATERIAL AND METHODS

In this study, a sample of 23 individuals was analyzed, which included: two males and one female of *Hamataliwa* sp, one male and three females of *Peucetia flava* Keyserling, 1877, nine males of *P. rubrolineata* Keyserling, 1877, and seven males of *Oxyopes salticus*

Hentz, 1845. All individuals were collected in Rio Claro (22°24'S, 47°33'W), State of São Paulo (SP), Brazil. The voucher specimens were deposited in the Laboratório de Artrópodes, Instituto Butantan (IBSP), city and State of São Paulo, Brazil. The chromosomal preparations were obtained according to the procedure described by Araujo et al. (2008). Chromosome spreads were stained with Giemsa solution (3% commercial Giemsa and 3% phosphate buffer, pH 6.8, in distilled water) for 15 min and subsequently silver-impregnated (Howell and Black, 1980) to detect the nucleolar organizer regions (NORs). Mitotic and meiotic nucleus records were performed using an Olympus BX51 microscope. The morphology of the chromosomes was determined according to the nomenclature proposed by Levan et al. (1964).

RESULTS

Mitotic metaphase cells of *Hamataliwa* sp stained with Giemsa showed the diploid numbers 2n = 28 for males and 2n = 30 for females, which were consistent with an X_1X_2/$X_1X_1X_2X_2$ sex chromosome system (Figure 1A). All chromosomes revealed telocentric morphology and decreased gradually in size. The sex chromosomes presented a high degree of condensation; the X_1 sex chromosome was slightly larger than the X_2 chromosome, but both possessed medium size. Early prophase I cells of male *Hamataliwa* sp revealed two blocks that were highly condensed and positively heteropycnotic, which were interpreted as sex chromosomes (Figure 1B). Diplotene and diakinesis nuclei showed 13II+X_1X_2 (Figure 1C,D), confirming the diploid number and type of sex chromosome system established through analyses of mitotic cells. In these latter meiosis-phases, the autosomal bivalents showed one interstitial or terminal chiasma and the X_1 and X_2 chromosomes always appeared as univalents and arranged side by side. In metaphase II cells, the haploid sets with n = 13+X_1X_2 and n = 13 chromosomes were observed (Figure 1E,F). All chromosome preparations of *Hamataliwa* sp were subjected to silver impregnation but only diplotene and diakinesis nuclei revealed NORs, which were located on the terminal region of one medium-sized bivalent and interstitial region of one small-sized bivalent (Figure 1G,H).

Giemsa-stained spermatogonial metaphases of *P. flava* and *P. rubrolineata* demonstrated the diploid chromosome numbers 2n = 22 and 2n = 21, respectively. The former species presented a sex chromosome system of the X_1X_2 type while the latter showed a system of the X type (Figure 2A,B). The karyotypes of both species were composed of telocentric chromosomes that gradually varied in size and sex chromosomes similar in size to the smallest elements of the diploid complement. In pachytene cells of *P. flava* and *P. rubrolineata*, the autosomal bivalents were fully synapsed and the sex chromosomes appeared as highly condensed and positively heteropycnotic univalents (Figure 2C,G). In diplotene and diakinesis nuclei, the meiotic formulas 10II+X_1X_2 for *P. flava* and 10II+X for *P. rubrolineata* were observed (Figure 2D,E,H,I). In both species, all autosomal bivalents showed one interstitial or terminal chiasma, with the exception of some cells of *P. flava* that presented one bivalent with two terminal and/or interstitial chiasmata. Metaphase II cells exhibited n = 10+X_1X_2 and n = 10 in *P. flava* (Figure 2F) and n = 10+X and n = 10 in *P. rubrolineata* (Figure 2J), confirming the regular segregation of all chromosomes during the preceding anaphase. Although the chromosome preparations of the two *Peucetia* species have been silver-impregnated, the presence of an argentophilic material corresponding to the NORs was not verified in the sample of cells examined.

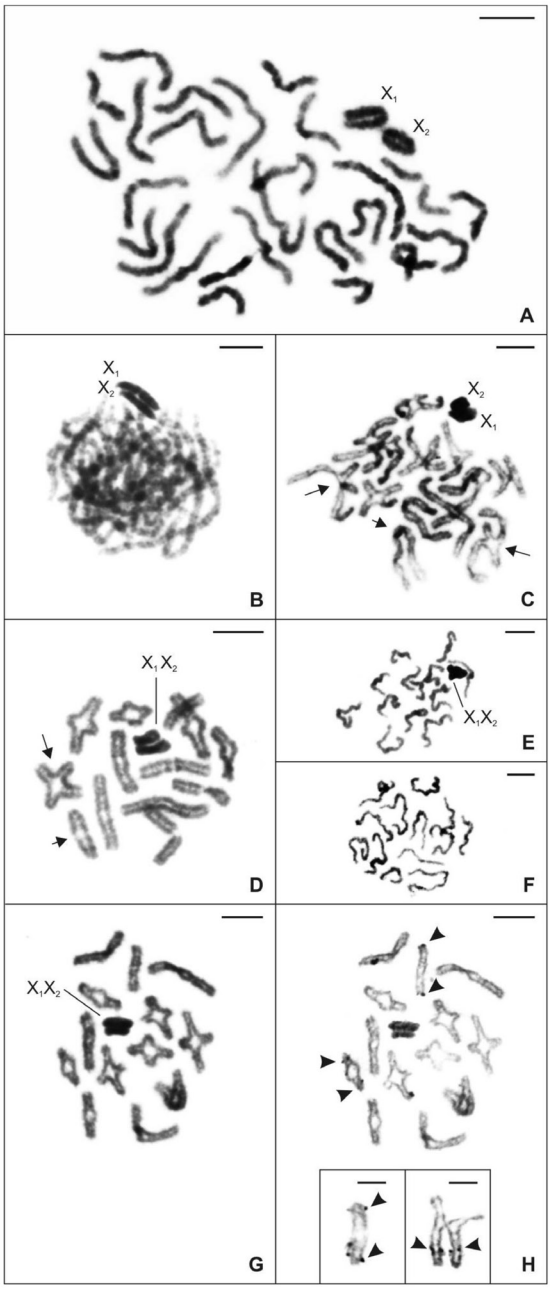

Figure 1. Testicular cells of *Hamataliwa* sp Giemsa-stained (A-G) and silver-impregnated (H). **A.** Mitotic metaphase with $2n = 26+X_1X_2$ and telocentric chromosomes. **B.** Pachytene, showing positively heteropycnotic sex chromosomes. **C. D.** Diplotene and diakinesis, exhibiting one interstitial (large arrows) or terminal (small arrow) chiasma per bivalent. **E. F.** Metaphase II nuclei with $n = 13+X_1X_2$ and n = 13, respectively. **G.** Diakinesis. **H.** The same cell as in G, revealing NORs (arrowheads) on terminal and interstitial regions of two bivalents. In detail, bivalents with one terminal and interstitial NORs. Scale bar = 10 µm (A-H) and 5 µm (detail in H).

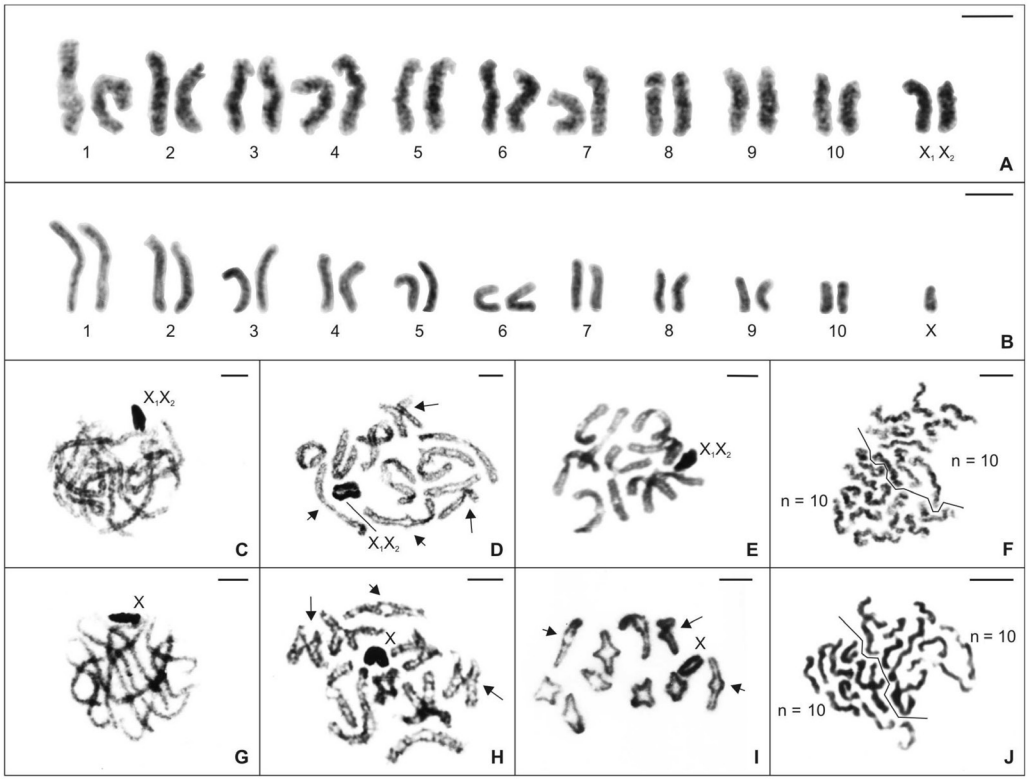

Figure 2. Testicular cells of *Peucetia flava* (A, C-F) and *Peucetia rubrolineata* (B, G-J) stained with Giemsa. **A. B.** Karyotypes, showing 2n = 20+X_1X_2 and 2n = 20+X, respectively. Note the telocentric morphology of all chromosomes. **C. G.** Pachytene with completely synapsed autosomal bivalents and highly condensed and stained sex chromosomes. **D. H.** Diplotene. **E. I.** Diakinesis. Large arrows point to interstitial chiasma and small arrows indicate terminal chiasma. **F. J.** Two cells in late metaphase II in which sister-chromatids are separated. Scale bar = 10 μm.

Mitotic metaphase cells of six male *O. salticus* after standard staining with Giemsa showed the diploid number 2n = 11, a sex chromosome system of the X type, and chromosomes with metacentric (pairs 1, 3 and 4), submetacentric (pair 2 and X-chromosome) and telocentric (pair 5) morphology (Figure 3A). In relation to the size, the chromosomes could be classified as large (pair 1), medium (pairs 2-4 and X-chromosome), and small (pair 5). Spermatogonial cells of one individual of the sample examined of *O. salticus* showed discrepant characteristics to those mentioned above (Figure 3B), i.e., the karyotype was composed of 2n = 12 chromosomes, including three heteromorphic chromosomes (one metacentric of large size and two telocentric of different sizes), four pairs of homomorphic chromosomes that correspond to pairs 2 to 5, and one submetacentric element identified as an X-chromosome. The three unpaired autosomes represented the heterozygous condition for the centric fusion between the chromosomes that originated the largest pair of the diploid complement. Moreover, in some cells of this specimen with 2n = 12, the presence of one extra chromosome possessing submetacentric morphology and extremely small size was detected (Figure 3C). This

element was interpreted as a B-chromosome. Thus, the spermatogonial cells of this individual exhibited 2n = 12 = 11+X or 2n = 13 = 11+X+B. Description of male meiosis of *O. salticus* was only based on the individuals that presented 2n = 11 chromosomes because in the chromosome preparations of the male with heteromorphic pair 1 and B-chromosome, most meiotic cells were in early prophase I stage. Pachytene nuclei exhibited 10 double filaments and one positively heteropycnotic block representative of the X-chromosome (Figure 3D). Diplotene cells revealed the meiotic formula 5II+X. Most autosomal bivalents presented a variable number of interstitial and/or terminal chiasmata from two to four, except the smallest bivalent that invariably showed one interstitial or terminal chiasma (Figure 3E). During prophase I, the X-chromosome was easily identified due to its high degree of condensation in relation to the

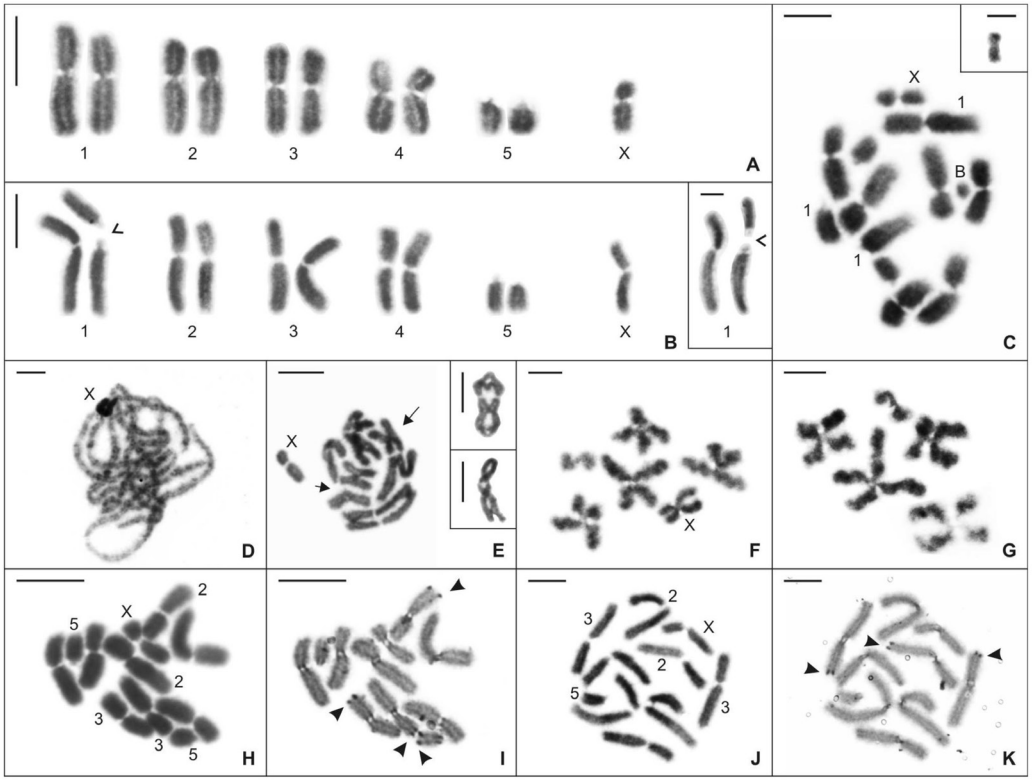

Figure 3. Testicular cells of *Oxyopes salticus* after staining with Giemsa (A-H, J) and silver impregnation (I, K). **A.** Karyotype with 2n = 10+X and metacentric, submetacentric, and telocentric chromosomes. **B.** Karyotype with 2n = 11+X. Note the heterozygous condition for the centric fusion between chromosomes that constitute pair 1. In detail, heteromorphic pair 1. **C.** Mitotic metaphase, 2n = 11+X+B, revealing the presence of one B-chromosome. In detail, the submetacentric B-chromosome. **D.** Pachytene. **E.** Diplotene, 2n = 5II+X, showing autosomal bivalents with interstitial (large arrow) and/or terminal (small arrow) chiasmata. In detail, bivalents with three and four chiasmata. **F. G.** Metaphase II cells with n = 5+X and n = 5, respectively. **H.** Mitotic metaphase, 2n = 10+X. **I.** The same cell as in H, revealing NORs (arrowheads) on the terminal region of pairs 2, 3 and 5. **J.** Incomplete mitotic metaphase with 2n = 10. **K.** The same cell as in J, exhibiting NORs (arrowheads) on the terminal region of pairs 2 and 3. Scale bar = 10 μm (A-K) and 5 μm (detail in B, C, E).

autosomes. Metaphase II spermatocytes showed two different haploid numbers, n = 5+X and n = 5 (Figure 3F,G). Silver-impregnated spermatogonial metaphases revealed six NORs on the long arm terminal region of pairs 2 and 5 and short arm terminal region of pair 3. However, only up to four NORs per cell were active (Figure 3H-K).

DISCUSSION

The family Oxyopidae possesses 430 species subdivided into nine genera (Platnick, 2010). Until now, a total of 21 species of the genera *Nishina*, *Oxyopes*, and *Peucetia* have been cytogenetically investigated. Among the four oxyopids studied here, only *O. salticus* has previously been examined (Painter, 1914). Additionally, this is the first chromosome record for a representative of the *Hamataliwa* genus. The $2n♂ = 26+X_1X_2$ with telocentric chromosomes observed in *Hamataliwa* sp was similar to the karyotype found in only one other species of this family, *P. viridana* (Stoliczka, 1869) (Bole-Gowda, 1950; Parida and Sharma, 1987; Sharma and Parida, 1987). It is worth pointing out that $2n♂ = 28$ represents the highest diploid number ever described for Oxyopidae. The karyotype characteristics verified in the two *Peucetia* species, $2n♂ = 20+X_1X_2$ in *P. flava* and $2n♂ = 20+X$ in *P. rubrolineata*, as well as the karyotype registered for *P. viridana*, $2n♂ = 26+X_1X_2$, revealed that in this genus there is a heterogeneity with regard to the diploid number and type of sex chromosome system. The telocentric chromosomal morphology encountered in the three species investigated here is, however, similar to that predominant in Oxyopidae spiders (Table 1). In relation to the size of the chromosomes, the only register in the literature refers to the sex chromosome of three species of the genus *Oxyopes*, in which the X-chromosome was classified as the largest or smallest element of the diploid complement. This interspecific discrepancy regarding the size of the X-chromosome could be related to variations in the quantity of constitutive heterochromatin and probably did not indicate an independent origin of this sex chromosome.

The results obtained in most individuals of the sample of *O. salticus* examined in the present study were surprising due to the occurrence of the diploid chromosome number $2n♂ = 11$. This diploid number is the lowest yet encountered in Oxyopidae and is the second lowest registered for Entelegynae spiders as a whole. The predominance of biarmed chromosomes is also very rare in entelegynes and has been verified in only 14 non-related species of a total of approximately 600 species analyzed cytogenetically (reviewed in Araujo, 2007). In addition, the chromosome characteristics of *O. salticus* from the Rio Claro population differed in relation to the karyotype already described for this same species, $2n♂ = 20+X_1X_2$, and for 18 other species of the genus *Oxyopes* that showed $2n♂ = 22+X$ and $2n♂ = 20+X$ (Table 1).

Within Oxyopidae, the great diversity of diploid number and type of sex chromosome system probably originated from the karyotype $2n♂ = 26+X_1X_2$ with acro/telocentric chromosomes. This karyotype certainly constitutes the basal type for this family, considering that it occurred in Senoculidae, a sister-group to Oxyopidae, and is commonly encountered in species of other families closely related to Oxyopidae, such as Trechaleidae, Lycosidae, and Pisauridae (Araujo, 2007; Giroti AM, unpublished results). Therefore, chromosomal rearrangement of the tandem fusion type between the autosomes was the mechanism responsible for the reduction of the diploid number, and tandem fusion between the X_1 and X_2 chromosome gave rise to the X-chromosome system. Moreover, the karyotypes observed in the individuals of *O. salticus* examined in this study revealed that centric fusions involving the autosomes

and the sex chromosomes also occurred during the karyotype evolution of Oxyopidae spiders. Král et al. (2006) analyzed certain species of Haplogynae spiders and also proposed that the X-chromosome system is derived from a multiple sex chromosome system. In contrast with the hypothesis formulated by us, i.e., the X-chromosome system arose by tandem or centric fusion between the X_1 and X_2 of the X_1X_2 system, Král et al. (2006) proposed that X system originated from the X_1X_2Y system. According to the authors, the conversion of the X_1X_2Y into an X system was a gradual process and the XY system was an intermediate stage.

The unusual karyotype $2n♂ = 10+X$, with a predominance of meta/submetacentric chromosomes, encountered in the sample of *O. salticus* studied here is certainly a derived type and evolved from $2n♂ = 20+X_1X_2$ described by Painter (1914) through "all or nothing" fusions as shown in Figure 4. We suggested that the karyotype of *O. salticus* from Rio Claro was originated by three main events: 1) centric fusion between eight autosomal pairs, producing four pairs of meta/submetacentric chromosomes; 2) centric fusion between the telocentric X_1 and X_2 chromosomes, converting the sex chromosome system into an X system; 3) tandem fusion between one ancestral autosomal pair of small size and the 1st derived autosomal pair, originating a chromosomal asymmetry in the derived karyotype due to the presence of one large and one small autosomal pair. Furthermore, the sympatric occurrence of one individual of *O. salticus* with the

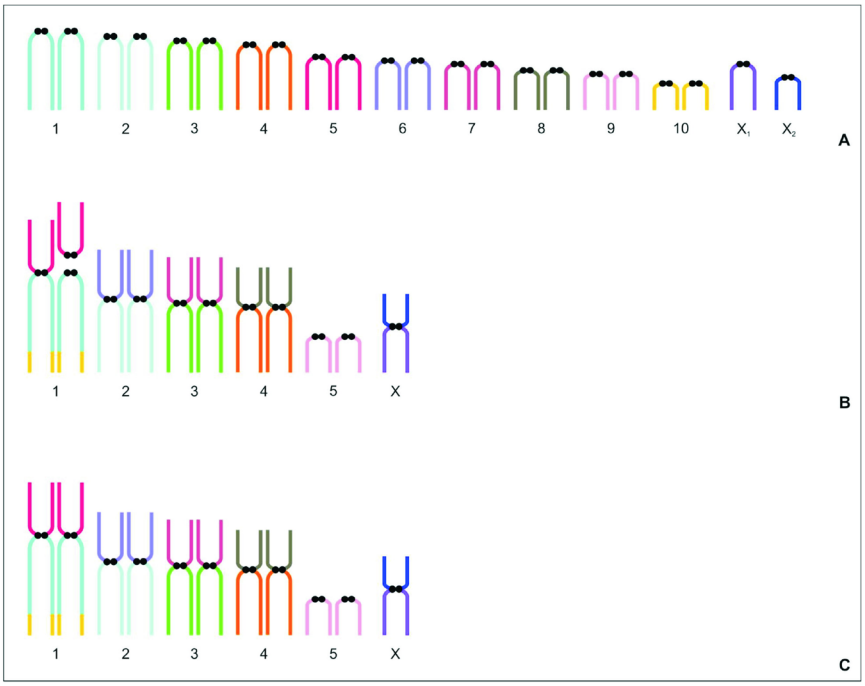

Figure 4. Schematic drawing showing a probable origin of the different chromosome constitutions observed in *Oxyopes salticus*. **A.** Ancestral karyotype with $2n♂ = 20+X_1X_2$ and telocentric chromosomes. **B.** Karyotype with $2n♂ = 11+X$ originated by centric fusion between 14 autosomal chromosomes and tandem fusion between one autosomal pair and the long arm of pair 1. A centric fusion between the X_1 and X_2 sex chromosomes converted the X_1X_2 sex chromosome system into an X type. Observe the heterozygous state of the centric fusion that originated the chromosomes of pair 1. **C.** Karyotype with $2n♂ = 10+X$, revealing the homozygous condition of the centric fusion of pair 1.

karyotype 2n♂ = 11+X, including the heterozygous state of the centric fusion that originated pair 1, revealed that the chromosome constitutions of *O. salticus* with all chromosomes in a fused state is not totally established in the population from Rio Claro. The chromosome characterization of a larger number of specimens from the population of Rio Claro and other localities would be very interesting and could reveal the presence of other karyotypes for this species.

Surprisingly, the diploid number variability in *O. salticus* was not only related to autosomal heteromorphism but also due to the presence of the B-chromosome. In spiders, the occurrence of the B-chromosome is extremely sporadic and has only been registered for six species of the families Amaurobiidae, Clubionidae, Lycosidae, Salticidae, Theridiidae, and Thomisidae (Montgomery, 1905; Painter, 1914; Avilés and Maddison, 1991; Rowell and Main, 1992; Qingtao et al., 1996). Unfortunately, in the specimen of *O. salticus*, carrier of the B-chromosome, the synaptic and segregational behavior of this additional chromosome was not established due to the fact that most meiotic cells were in early prophase I stage.

The analysis of meiotic cells of *Hamataliwa* sp, *P. flava*, *P. rubrolineata*, and *O. salticus* permitted us to confirm the diploid number, chromosomal morphology, and the main type of sex chromosome system, established through the study of mitotic cells in the four species. Additionally, the investigation of diplotene and diakinesis nuclei showed that one chiasma per bivalent, frequently observed in other oxyopids, also occurred in *Hamataliwa* sp, *P. flava*, and *P. rubrolineata*. In contrast, the high number of chiasmata per bivalent was verified in all large-sized bivalents of *O. salticus*. Similarly to this latter species, the presence of more than one chiasma per bivalent was also observed in some beetles whose diploid complement seems to have evolved by fusions (Schneider et al., 2007). According to John (1990) the number of chiasma per bivalent is partially dependent on the chromosome size. In *O. salticus*, a clear relationship between chromosome size and the number of chiasma per bivalent was noticed, since the small bivalent invariably exhibited only one interstitial or terminal chiasma.

The information on NOR in spiders is very scarce. Within Entelegynae the analysis of this specific chromosome region was performed only in certain species, e.g., one species of Lycosidae, Nephillidae, Oxyopidae, and three species of Sparassidae, which revealed NORs on the terminal region of one, two, or up to three autosomal pairs (Wise, 1983; Barrion et al., 1989; Araujo et al., 2005; Rodríguez-Gil et al., 2007). Although *Hamataliwa* sp and *O. salticus* differed with regard to the number of carrier pairs of NOR, these two species always presented argentophilic material on autosomal chromosomes. The occurrence of NOR only on autosomal chromosomes could be a shared characteristic for Entelegynae, unlike Haplogynae, in which Oliveira et al. (2007) proposed that NORs on both autosomes and sex chromosomes seem to be the ancestral pattern for the group. Nevertheless, the presence of NOR on sex chromosomes of some entelegyne spiders cannot be excluded, considering the low number of species examined in this manner. In *Hamataliwa* sp, NOR on the interstitial region of one autosomal pair could also represent a derived pattern, taking into account that in most spiders and animal species the NOR is frequently located on the terminal region (Sumner, 2003). The change in position of the nucleolar organizer site in *Hamataliwa* sp could be attributed to chromosome rearrangements of the translocation or inversion type.

The analysis of the four Oxyopidae spiders and data available in the literature permitted us to suggest the trends in chromosome evolution for this family. Although certain species retain the ancestral chromosome constitutions 2n♂ = 26+X_1X_2 with acro/telocentric chromosomes, the vast majority of the oxyopids have their karyotype differentiated by both reduction

in diploid chromosome number and change of the sex chromosome system to X type. These mechanisms of chromosome evolution resulted in an interspecific and intraspecific karyotype variability within the family Oxyopidae. The most remarkable karyotype differentiation occurred in the specimens of *O. salticus* studied here, which showed one of the lowest diploid numbers ever recorded for Entelegynae spiders. The use of techniques to highlight specific chromosome regions in a large number of species will certainly reveal more details of chromosome evolution in oxyopids.

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