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CHROMOSOME STUDIES IN THE GENUS *PTEROTRICHA* (GNAPHOSIDAE) FROM TURKEY

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Abstract - The diploid chromosome set and male meiosis of two spider species belonging to the family Gnaphosidae from Turkey are described. Both *Pterotricha kochi* and *Pterotricha lesserti* exhibited the diploid chromosome number as 2n♂=22 and X_1X_20 sex chromosome type. All chromosomes were acrocentric. During the first meiotic division, both species had ten autosomal bivalents and two sex chromosomes that were positively heteropycnotic. At the second meiotic division, two kinds of nuclei were determined, with or without sex chromosome. These data will contribute to our knowledge of the evolution of the genus *Pterotricha*.

Keywords: karyotype; meiosis; sex chromosome system; Araneae

INTRODUCTION

Gnaphosidae, a family of the suborder Araneomorphae, comprises 121 genera distributed worldwide (Platnick, 2013). In Turkey it is represented by 30 genera and 132 species, four of them belong to the genus *Pterotricha* Kulczynski, 1903 (Bayram et al., 2013).

Several studies of the cytogenetic aspects of spiders have been conducted. To date, 706 species of spiders belonging to 65 families and 275 genera have been studied (Araujo et al., 2013). Based on these studies, araneomorph spiders display a constant acrocentric morphology of chromosomes in many species. Also a notable feature of karyotypes is the predominance of unusual multiple X chromosome systems such as ♂X_1X_2/♀$X_1X_1X_2X_2$, often assigned as X_1X_20, where the 0 indicates the absence of the Y chromosome (Král et al., 2006).

In this study, the karyotypes and meiotic courses of *Pterotricha kochi* (O. P.-Cambridge, 1872) and *Pterotricha lesserti* Dalmas, 1921, were analyzed by conventional Giemsa staining for the first time. Karyological analysis has proved to be useful in determining the cytogenetic relationship and evolution of different species.

MATERIALS AND METHODS

Adult specimens of *P. kochi* and *P. lesserti* (Gnaphosidae) were collected from natural populations between March and June 2012 and transferred alive to the laboratory. A sample containing 21 individuals was used; these were *P. kochi* – 12 adult males from Sakçagözü, Gaziantep (37°08′37.94″N, 36°52′47.92″E); Zorkun, Osmaniye (37°01′55.71″N, 36°18′54.15″E); Yayladağ, Hatay (35°53′48.64″N, 36°03′49.97″E), and *P. lesserti* – 9 adult males from Islahiye, Gaziantep (37°01′20.56″N, 36°37′26.26″E

), Kâhta, Adıyaman (37°48′18.69″N, 38°36′53.43″E). The specimens were deposited in the arachnological collection after dissection in Nevşehir University, Art and Science Faculty, Department of Molecular Biology and Genetics, Nevşehir, Turkey.

Dissection of spiders and methods of chromosome preparations were performed according to Traut (1976) with a modification for hypotonisation time (12-15 min). The gonads were fixed by two changes of Carnoy fixative that contained ethanol, chloroform and glacial acetic acid (6:3:1), respectively. A piece of tissue was suspended in a drop of 60% acetic acid on a slide using a pair of tungsten needles. The slide was dried on a histological plate (surface temperature 42°C) and stained with a 5% Giemsa solution in Sorensen phosphate buffer (pH 6.8) for 27 min at room temperature. Mitotic and meiotic cells were determined under Olympus BX53 microscope and photographed using a DP26 digital camera (Olympus) using CellSens software (Olympus). Karyotypes were performed by arranging chromosomes in pairs using calculations of relative chromosome lengths with sex chromosomes (X_1 and X_2) from 10 spermatogonial metaphases by CellSens software. The chromosomes were identified based on centromeric position according to Levan et al. (1964).

RESULTS

Karyotypes and sex chromosome systems

Karyotypes of the two *Pterotricha* species studied consisted of 22 acrocentric chromosomes that decrease gradually in size (Fig. 1.A and B). The chromosome lengths of autosome pairs ranged from 8.06% to 5.12% in *P. kochi* and from 8.36% to 5.62% in *P. lesserti*. The sex chromosome system was X_1X_20 in males for both *P. kochi* and *P. lesserti*. Chromosome lengths of X_1 and X_2 were 6.46% and 5.82% in *P. kochi*; 6.74% and 5.90% in *P. lesserti*, respectively. The sex chromosomes showed approximately the similar size.

Meiosis

The principal steps of male meiosis were observed. Between leptotene to pachytene the sex chromosomes resided along the nuclear periphery and were observed as a positively heteropycnotic material (Figs. 2A and D). Diplotene, diakinesis and metaphase I showed 10 bivalents and 2 univalent sex chromosomes (Figs. 2B and E). Sex chromosomes X_1 and X_2 were easily detected at the first prophase and metaphase stages due to their strong heteropycnotic behavior. During the second meiotic stages, two types of nuclei, either with (n=12) or without (n=10) sex chromosomes were obtained. The sex chromosomes were positively heteropycnotic and clearly distinct from the second prophase and metaphase in *P. kochi* (Fig. 2C), however, X_1 and X_2 could not be distinguished because of their isopycnotic behavior in *P. lesserti* (Fig. 2F).

DISCUSSION

The diploid chromosome number among the different genera of the family Gnaphosidae varies from 21 to 30 in males, with a remarkable predominance of 2n♂=22. Exceptions to this diploid number are observed in *Urozelotes rusticus* (L. Koch, 1872) (2n♂=21; Srivastava & Shukla, 1986), *Nodocion floridanus* (Banks, 1896) (2n♂=24; Tugmon et al., 1990), *Scotophaeus blackwalli* (Thorell, 1871) (2n♂=24; Mittal, 1961; Mittal, 1967) and *Scotophaeus domesticus* Tikader, 1962 (2n♂=30; Srivastava and Shukla, 1986). Despite these exceptions, 2n♂=22 is suggested to be the ancestral karyotype for gnaphosids.

The sex chromosome system in gnaphosids seems to be uniformly X_1X_20 in males, except in *U. rusticus* and two undetermined *Drassodes* Westring 1851 species (Srivastava and Shukla, 1986), which exhibit an X0 system. This acrocentric X chromosome probably originated through the gradual elimination of one X chromosome of X_1X_2 type or by reciprocal translocation between X_1 and X_2 chromosomes that was preceded by distal fission in one sex chromosome and proximal fission in the other X (Maddison and Leduc-Robert, 2013). Moreover, the chromosome morphology of this family in males is acrocentric, except for *Gnaphosa sp*. (Datta and Chatterjee, 1989), which shows telocentric chromosomes.

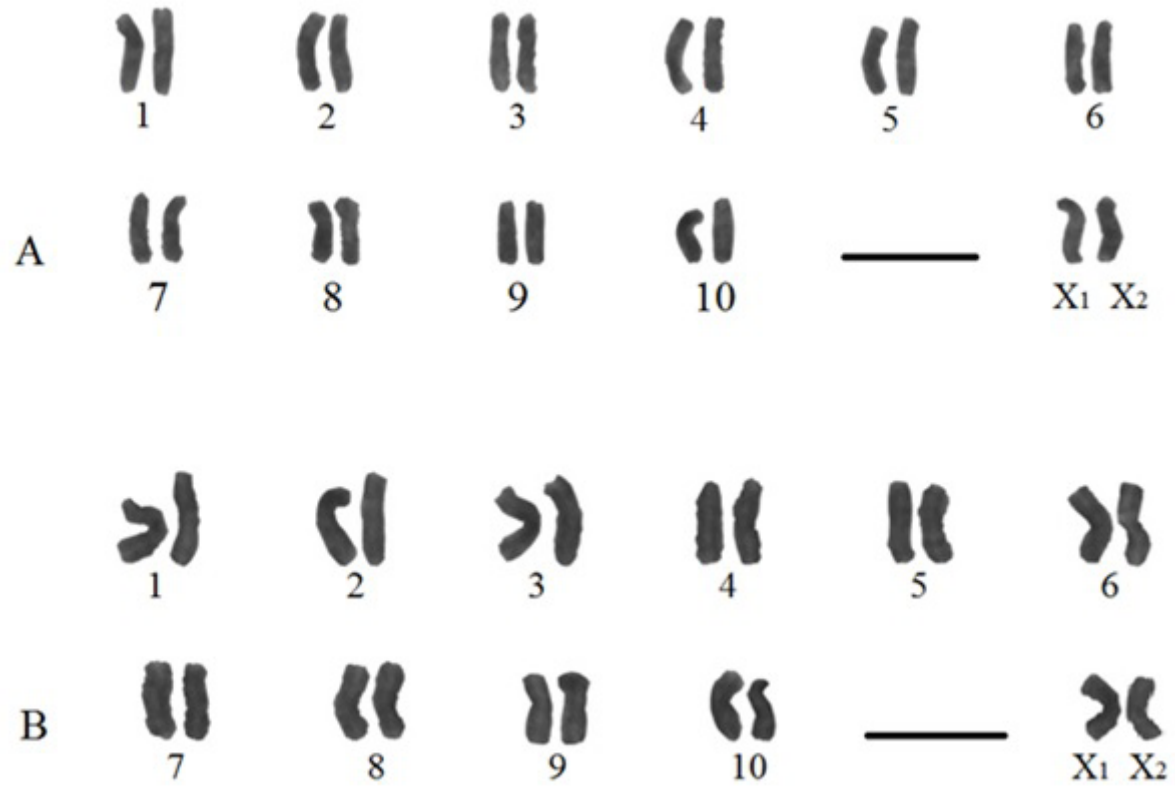

Fig. 1. Karyotypes of (A) *Pterotricha kochi* (B) *Pterotricha lesserti* (Scale= 10 μm)

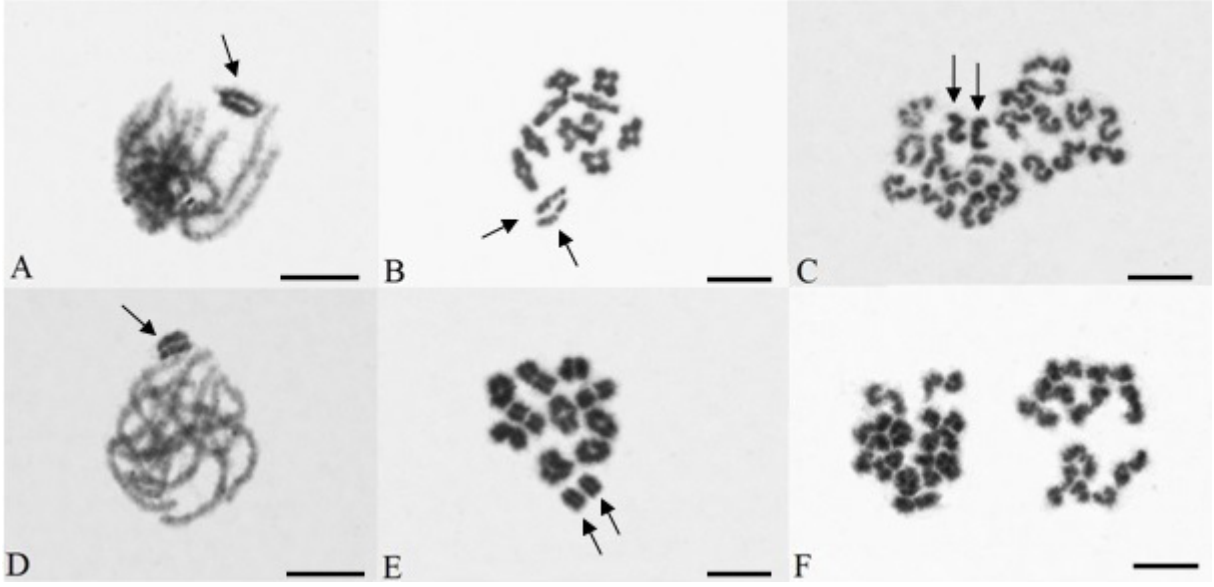

Fig. 2. *Pterotricha kochi* (A) Pachytene (B) Diplotene (C) Prometaphase II, *Pterotricha lesserti* (D) Pachytene (E) Diplotene (F) Prometaphase II (arrows indicate sex chromosomes, Scale= 10 μm)

To date, two species of the genus *Pterotricha* have been analyzed: the diploid numbers and sex chromosome systems have been determined as 2n♂=22, X_1X_2 in *Pterotricha dalmasi* Fage, 1929 and *Pterotricha procera* (O.P.-Cambridge, 1874) (Gorlova et al., 1997). In this study, the results of *P. kochi* and *P. lesserti* are similar to the results of Gorlova et al. (1997). It seems that the chromosome number and sex chromosome system are relatively conservative in the genus *Pterotricha*.

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