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Chromosomal Study on three Araneids Spiders from Akola District

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Abstract
The cytologenetic studies on three spider species of Genus *Cyrtophora citricola, Neoscona nautica,* and *Eriovixia excelsa* were studied based on the samples collected in and around Akola District. The gonads were dissected out and metaphasic chromosome preparation was made using standard staining techniques. The chromosomes diploid number (2n) and sex chromosomes system in females of all species were found in the same, 2n=24, (24 + X_1X_20) , and all are telocentric, as all chromosomes shows, terminal centromere.
Keywords: Araneidae, Sex chromosome, *Cyrtophora citricola, Neoscona nautica, Eriovixia excelsa,* 2n = 24

Introduction:

Spiders belong to e the largest order Araneae in class Arachnida and rank seventh in total species diversity among all other orders of animals. (Sebastian and Peter, 2009). As of November 2015, at least 45,700 spider species, and 113 families have been recorded by taxonomists (World catalog 2016). Uptill now only one percentage of cytologenetic studies on spiders has been reported. Carnoy (1885) presented the first, although inaccurate, chromosome numbers of some spider species. However, this study did not mention the existence of chromosomes that could be related to sex determination. Araneidae (orb-weaving spiders) are known to have XO (male)-XX (female) and X1X2O (male)-X1X1X2X2 (female) sex determination mechanisms (Mittal 1960; Datta and Chatterjee 1988). While most araneid species possess the X1X2O-X1X1X2X 2 mechanisms, there are a few that have XO-XX sex determination (Datta and Chatterjee 1988). The study of spiders Karyotypes is important for comparison with different species that allows us, to determine their taxonomic position, and also useful to study phylogeny.

The first cytogenetic study in spiders performed by Carnoy (1885), in his experiment he imbedded female or male gonads in paraffin, sectioned and stained with Heidenhein's iron Haematoxylin. Decades later, Sharma *et al.,* (1959) and Beçak and Beçak (1960) observed spider chromosome by aceto-orcein or aceto carcin method. Pinter and Walters (1971) introduced the use of colchicine solution for cytological preparations of spider testes and ovaries. In the same decade, Brum-Zorrila and Cazenave (1974) applied 3:1 methanol:acetic acid as a fixative solution and Giemsa solution as a stain. Spider shows multiple sex chromosome sex ♂/X1X2/♀/X1X1X2X2 (Benavente *et al.,* 1982).

This system is referred as X1X20 (where 0 denotes the absence of the chromosome Y). This system is found in 77% of the spider species studied so far(Araujo *et al.,* 2005). Such determination is probably ancestral in spiders as it was found also in the most plesiomorphic recent spider taxon, the Mesothelae (Suzuki 1954). The X1X2X30, X1X2X3X40 and X0 systems were originated from X1X20 system. These systems are evolved by non-disjunction of X chromosome (Brum-Zorrilla and Postiglioni 1981), the last one was derived mostly by centric or tandem fusion of the original sex chromosomes X1 and X2 (Kral 1994). In this paper, we cytogenetically analyze the diploid number, chromosome morphology and sex determining mechanism of 3 species of araneid spider species *Cyrtophora citricola, Neosconanautica,* and *Eriovixia excelsa* in and around Akola district.

Materials and Methods

Cyrtophora citricola, Neoscona nautica, and *Eriovixia excelsa*, spiders were collected in and around Akola district from September to January between 2011 and 2012. All specimens were alive and were moved to the laboratory. They were identified with the help of available keys. The abdomen of spider dissected under stereo microscope and Gonads (ovaries from female) were removed. For cytogenetic preparations, the protocol given by Sergio Gustavo Rodrı´guez-Gil (2007) was followed with some modification .The specimen were injected with 0.1 ml of 0.01% colchicine solution. After 1hr, gonads were dissected by removal of other tissues on the cleaned slide. Each sample was dispersed in 2 ml of hypotonic solution (KCl 0.56%) for 10-15 min, centrifuged at 400 rpm for 10 min, and fixed in 1 ml of 3 : 1 (methanol : acetic acid) for 30 min . The cell suspension was dropped onto clean slides in such a way that the drops should not be overlapped, air-dried and stained with Giemsa 3% for 10-15 min then rinsed in distilled water to remove excess Giemsa

stain and mount with DPXfor chromosome counts and karyotyping..

Chromosome spread slides were observed under Magnus MLX-B Plus Binocular research Microscope with 40X ocular and 100X objectives. and the best metaphase pictures were photographed with a digital camera system attached to the microscope. The chromosome groups were determined on the slides and their films were photographed; every chromosome was drove in a circle and Diploid chromosome numbers were obtained after counting circles. Karyotypes of all three species of spiders were constructed by arranging chromosomes in pairs according to size using reported images. Chromosome lengths (RCLs) of each chromosome pair was determined from 10 metaphase plates obtained from each species. Chromosome morphology was classified according to the method of Levan *et al.,* (1964).

Results and Discussion

Figs. 1, 2 and 3 and Table, 1 depicted the oogonial metaphase count of *Cyrtophora citricola,(Forskal),* Neoscona*nautica,* (Simon) and *Eriovixia excelsa*from Araneidae family. The chromosomes in mitotic stages appear to be small and rod-shaped.

In *Cyrtophora citricola,(Forskal)* the female diploid chromosome number 2n is 24 (Figure 1). All autosome pairs were telocentric. Autosome pairs with gradually reducing size. The sex chromosome system was of the X_1X_2X0type. The X_1 and X_2 sex chromosomes were also telocentric. Contrasting to our reported 2n=24 chromosomes, for *Cyrtophora citricola,* Datta, and Chatterjee. (1988) reported as 26 with Chromosome sex system as $X1X1X_2X_2$, however all chromosomes were reported to be are telocentric.

In *Neoscona nautica* female diploid chromosome number 2n is 24. (Fig2). All autosome pairs were telocentric and are relative in size. The sex chromosome system was of the X_1X_20type. The X_1 and X_2 sex chromosomes were also telocentric. The results are similar with that of Parida and Sharma (1987), who too reported 2n as 24 chromosome and chromosome sex systems as X_1X_20 in different Neoscona genera from India.

Table. 1-Chromosomal details of Cyrtophora citricola, Neoscona nautica, and Eriovixia excelsa, spiders from Akola district.

Species	Sex	No of individuals studied	2n	n	Sex determination
Cyrtophora citricola,	F	*9*	*24*	11 13	X_1X_2X0
Neoscona nautica,	F	8	24	11 13	X_1X_2X0
Eriovixia excelsa,	F	10	24	11 13	X_1X_2X0

In *Eriovixia excelsa*female diploid chromosome number 2n is 24 (Fig 3). All autosome pairs were telocentric and are gradually decreasing in size. The sex chromosome system was of the X_1X_20type. The X1 and X2 sex chromosomes were also telocentric.Parida and Sharma (1987) and Datta, and Chatterjee. (1988) found and reported similar karyology forspiders of genus Eriovixia from India, they documented that chromosome morphology was *as 22T*+XX1 2T and the chromosome sex systems as X_1X_2.

Thus this study, concluded that, all three studied species *Cyrtophora citricola,(Forskal),* Neoscona*nautica,* (Simon) and *Eriovixia excelsa*from Araneidae family collected from Akola district, have similar 2n = 24 chromosomes, while observed sex chromosome complement were also common in them as X_1X_20type.

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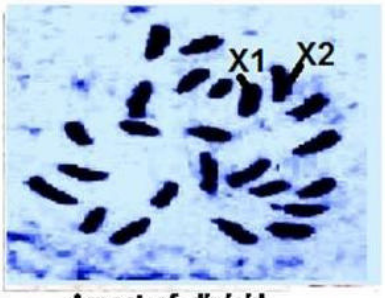
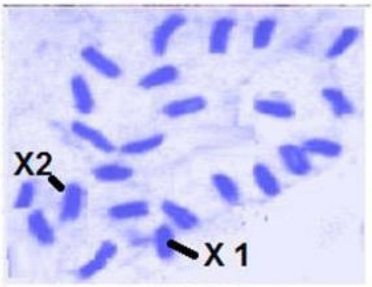
Fig.1. *Cyrtophora citricola, Female*



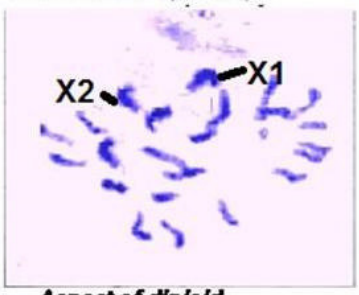
Aspect of diploid chromosomes (2n=24).

Fig.2. *Neoscona nautica,* Female



Aspect of diploid chromosomes (2n=24).

Fig.3. *Eriovixia excelsa,* female



Aspect of diploid chromosomes (2n=24).

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