

Cytogenetic characters of *Arachnocoris trinitatus* Bergroth, 1916 (Insecta: Heteroptera: Nabidae) from nests of the spider *Coryssocnemis simla* Huber, 2000 (Araneae: Pholcidae)

V.G. Kuznetsova¹, S. Grozeva²

¹Zoological Institute, Russian Academy of Sciences, Universitetskaya nab. 1, St. Petersburg 199034, Russia; ²Institute of Zoology, Bulgarian Academy of Sciences, Blvd Tsar Osvoboditel 1, Sofia 1000, Bulgaria.

E-mails: karyo@zin.ru, sgrozeva@yahoo.com

Abstract. The study of karyotype and testis structure has been carried out on males of the nabid bug *Arachnocoris trinitatus* Bergroth, 1916 collected from web nests of the pholcid spider *Coryssocnemis simla* Huber, 2000 in Trinidad, West Indies. Testes were shown to consist each of 3 follicles. The chromosome complement includes $2n = 12$ (10 + XY). Nucleolus organizing regions (NORs) are GC-rich and are situated on the largest autosome pair. These characters match those of *A. trinitatus* bugs inhabiting nests of the other pholcid species *Mesabolivar aurantiacus* Mello-Leitão, 1930 in Trinidad.

Key words: Araneae, Pholcidae, *Coryssocnemis simla*, Heteroptera, Nabidae, *Arachnocoris trinitatus*, follicle number, karyotype, DAPI, CMA₃.

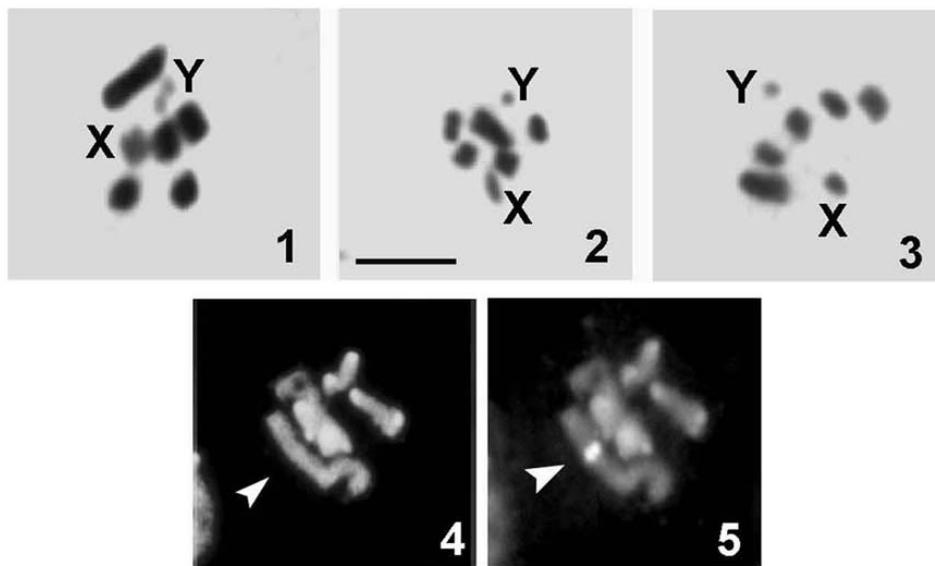
INTRODUCTION

The bug tribe Arachnocorini (Nabidae, Nabinae) occurs in the New World and represents a morphologically and biologically highly specialized group of damsel bugs, with only two genera, *Pararachnocoris* Reuter, 1908 and *Arachnocoris* Scott, 1881 (Kerzhner, 1981). The small neotropic genus *Arachnocoris* includes eleven species (Kerzhner, 1986, 1990), all of which inhabit spider web nests, mainly of pholcid spiders (Araneae, Pholcidae).

In Trinidad (West Indies), *Arachnocoris trinitatus* (Bergroth, 1916) is known as an endemic and a frequent species in web nests of the pholcid spider *Mesabolivar aurantiacus* Mello-Leitão, 1930 (Sewlal, Starr, 2008). Recently, Sewlal and Starr (unpublished) collected for the first time *Arachnocoris* bugs

in Trinidad in web nests of the other pholcid spider *Coryssocnemis simla* Huber, 2000. The bugs were identified by Dr. Tom Henry as *A. trinitatus*. It is the authors' opinion that *Arachnocoris* bugs use spider nests as ready-made prey-capture devices and as the sites to find mates (Sewlal, Starr, 2008).

Quite recently, as a part of ongoing studies of the family Nabidae, we examined male and female reproductive organs and karyotype of *A. trinitatus*, collected in Trinidad in *M. aurantiacus* nests (Kuznetsova et al., 2007). In that study, *A. trinitatus* was found to display a number of characters differentiating it from all hitherto studied nabid species (Kuznetsova et al., 2004; Grozeva et al., 2004). Among these characters are chromosome number, $2n = 12$ (10 + XY), the lowest within the family; the



Figs 1-5. Chromosomes of *Arachnocoris trinitatus*, male, $2n = 10 + XY$. **1** - metaphase I. The X and Y chromosomes are clearly double-stranded. **2** - metaphase II viewed from the pole. The X and Y chromosomes are widely separated and clearly single-stranded. **3** - metaphase II viewed from the side. The X and Y chromosomes demonstrate a "distance pairing". **4** - DAPI-stained condensation stage with AT-rich blocks at telomeres. Noteworthy is the absence of signals on the largest bivalent (arrow). **5** - the same chromosome plate after CMA_3 . Noteworthy is the bright interstitial GC-signal on the largest bivalent (arrow). Bar = $10\mu m$.

nucleolus organizing regions (NORs, i.e. sites of the RNA genes) situated on the autosomes rather than on the sex chromosomes, which is characteristic of all other representatives of the family; testes composed of 3 seminal follicles but not of 7, the modal number for the Nabidae. These distinctive characters, at least the reduced follicle number, were suggested to indicate many transformations during speciation of *A. trinitatus* and probably represent the result of extreme specialization of the species. In order to characterize the karyotype of *A. trinitatus* in greater detail, we used, in addition to the standard Schiff-Giemsa staining, C-banding and silver nitrate staining (NOR-banding), also base-specific fluorochrome staining (CMA_3 and DAPI) for revealing GC- and AT-rich chromosome sites.

The goal of the present study was to check if *A. trinitatus* inhabiting *C. simla* nests dis-

plays any characters different from those found previously in *A. trinitatus* from *M. aurantiacus* nests. For this purpose, the same set of cytogenetic techniques has been used.

MATERIAL AND METHODS

Males of *A. trinitatus* were collected in web nests of *C. simla* spiders. The collection was made by J.N. Sewlal and Ch.K. Starr (unpublished) in the higher elevations of the deciduous seasonal forest above the Mt. St. Benedict monastery in Trinidad, West Indies ($1180079N$, $674919E$) in April 2008 (late dry season).

A total of 7 males were studied, and all chromosome preparations were made from testes. We applied here the same cytogenetic techniques as in Kuznetsova et al. (2004).

The preparations were analyzed by an

Olympus BX 51 light microscope at 1000x, and photomicrographs taken using a digital camera MotiCAM 2000. The fluorochrome-labeled preparations were analyzed using a fluorescence microscope Dialux 22 at 1000x, and photomicrographs were taken using a camera Wild Leitz MPS 46.

RESULTS AND DISCUSSION

The testes consist of three elongated colorless follicles (testis tubes). In spermatocyte prophase I, the homologues of the bivalents are oriented parallel to each other, and the diplotene and diakinesis stages are absent, which is characteristic of the achiasmatic meiosis. This meiotic pattern is common to the Nabidae as a whole (Nokkala, Nokkala, 1884) and agrees with that of *A. trinitatus* males inhabiting *M. aurantiacus* nests (Kuznetsova et al., 2004). The spermatocyte metaphases I (MI) contain 5 autosomal bivalents and the X and Y univalents (Fig. 1) confirming the meiotic formula of $n = 5A$ (autosomes) + X + Y and the diploid chromosome complement of $2n = 12$ (10 + XY). The chromosomes are holokinetic displaying no localized centromeres. At this stage, the X and Y chromosomes are clearly double-stranded. The longest bivalent appears to be more than two times as large as the rest bivalents, which have similar sizes. The first division is reductional for the bivalents, contrasting with the sex chromosomes, which undergo equational separation in anaphase I (AI) and segregation in anaphase II (AII). This pattern known as a sex chromosome post-reduction is characteristic of the overwhelming majority of the Heteroptera (Ueshima, 1979), the family Nabidae included (Nokkala, Nokkala, 1884; Kuznetsova, Maryńska-Nadachowska, 2000). Fig. 2 and Fig. 3 show metaphase II cells (MII) each with 5 double-stranded autosomes and the single-stranded X and Y chromosomes. Viewed from the side (Fig. 3), sex

chromosomes demonstrate a bipolar co-orientation and are located near the opposite poles. This pattern named a “distance-pairing” is typical of the subfamily Nabinae as a whole (Kuznetsova et al., 2004, 2007; Angus et al., 2008).

After DAPI and CMA₃ treatment, prophase condensation stages showed the distribution of signals similar to that previously discovered in *A. trinitatus* from *M. aurantiacus* nests. After DAPI, different in size terminal AT-rich blocks were visible in the majority of bivalents except for the largest bivalent, which was fully DAPI-negative (Fig. 4). When stained by CMA₃, the same chromosome plate showed terminal blocks on the same bivalents and a bright interstitial GC-rich block, the site of NOR, on the largest bivalent (Fig. 5).

In conclusion, we have found that *A. trinitatus* bugs inhabiting *C. simla* web nests share with those inhabiting *M. aurantiacus* web nests the same testis structure and similar cytogenetic characters.

ACKNOWLEDGMENTS

We are grateful to Drs J.N. Sewlal and Ch.K. Starr for providing the fixed insects for the present study. This study was supported financially (for V.G. Kuznetsova) by the Russian Foundation for Basic Research (grant 08-04-00787), the Programs of the Presidium of the Russian Academy of Sciences “Dynamics of Gene Pools in Animals, Plants and Man” and “The Origin and Evolution of Biosphere”, and (for S. Grozeva) by the Bulgarian Academy of Sciences and the Bulgarian Ministry of Education and Science (grant TK-B-1601).

REFERENCES

- Angus R.B., McCartney S., Thompson L. 2008. On the karyotypes of the British high chromosome number Nabidae (Insecta: Heteroptera) // *Comp. Cytogenet.* 2(1): 67-72.

- Grozeva S., Kuznetsova V., Nokkala S. 2004.** Patterns of chromosome banding in four nabid species (Heteroptera, Cimicomorpha, Nabidae) with high chromosome number karyotypes // *Hereditas*. 140: 99-104.
- Kerzhner I.M. 1981.** Nasekomye khobotnye (Insecta: Rhynchota). Nabidae. *Fauna SSSR*. 13(2). Leningrad. 326 p. (In Russian).
- Kerzhner IM 1986.** Neotropical Nabidae (Heteroptera), 1: A new genus, some new species, and notes on synonymy // *J. New York Entomol. Soc.* 94(2): 180-193.
- Kerzhner I.M. 1990.** Neotropical Nabidae (Heteroptera), 3: Species of the genus *Arachnocoris* from Costa Rica // *J. New York Entomol. Soc.* 98(2): 133-138.
- Kerzhner I.M. 1996.** Family Nabidae A. Costa 1983 – damsel bugs, (pp. 84-107) // Aukema B., Rieger C. (Eds). *Catalogue of the Heteroptera of the Palaearctic Region*. 2. Amsterdam. 361 p.
- Kuznetsova V.G., Maryańska-Nadachowska A. 2000.** Autosomal polyploidy and male meiotic pattern in the bug family Nabidae // *J. Zool. Syst. Evol. Res.* 38: 87-94.
- Kuznetsova V., Grozeva S., Nokkala S. 2004.** New cytogenetic data on Nabidae (Heteroptera, Cimicomorpha), with discussion on karyotype variation and meiotic patterns and their taxonomic significance // *European J. Entomol.* 101: 205-210.
- Kuznetsova V.G., Grozeva S., Sewlal J. N., Nokkala S. 2007.** Cytogenetic characterization of the endemic of Trinidad, *Arachnocoris trinitatus* Bergroth: the first data for the tribe Arachnocorini (Heteroptera: Cimicomorpha: Nabidae) // *Folia Biol. (Krakow)*. 55(1-2): 17-26.
- Nokkala S., Nokkala C. 1984.** Achiasmatic male meiosis in the heteropteran genus *Nabis* (Nabidae, Hemiptera) // *Hereditas*. 101: 31-35.
- Sewlal J.N., Starr C.K. 2008.** Observations of the insect *Arachnocoris trinitatus* (Heteroptera: Nabidae) as an inquiline in the webs of the spider *Mesabolivar aurantiacus* (Araneae: Pholcidae) // *Caribbean J. Sci.* 44(1): 132-135.
- Ueshima N. 1979.** Hemiptera II: Heteroptera. *Animal Cytogenetics*. 3: *Insecta* 6. Berlin-Stuttgart. 113 p.

Received November 20, 2008.

Accepted by I.A. Gavrilov, December 20, 2008.

Published December 30, 2008.