

Cytogenetic features of some grasshoppers (Orthoptera, Pamphagidae and Acrididae) from Israel

Цитогенетические особенности некоторых саранчовых (Orthoptera, Pamphagidae и Acrididae) Израиля

A.G. Bugrov, O.G. Buleu
А.Г. Бугров, О.Г. Булэу

Institute of Systematics and Ecology of Animals, Russian Academy of Sciences, Siberian Branch, Frunze Str. 11, 630091 Novosibirsk, Russia; Novosibirsk State University, Pirogova Str. 2, Novosibirsk 630090, Russia. E-mail: bugrov04@yahoo.co.uk, bugrov@fen.nsu.ru.

Институт систематики и экологии животных СО РАН, ул. Фрунзе 11, Новосибирск 630091 Россия; Новосибирский госуниверситет, ул. Пирогова 2. Новосибирск 630090 Россия.

Key words: Pamphagidae, Acrididae, grasshoppers of Israel, karyotypes, autosomes, sex chromosomes, C-banding.

Ключевые слова: Pamphagidae, Acrididae, саранчовые Израиля, кариотипы, аутосомы, половые хромосомы, С-бэндинг.

Abstract. For the first time the cytogenetic features of some grasshopper species from Israel were studied. In this paper we conducted comparative cytogenetic analysis of four Israel grasshoppers belonging to Pamphagidae (*Tmethis pulchripennis* (Serville, 1838) and *Prionosthenus syriacus* (Brisout de Barneville, 1854)) and Acrididae (*Heteracris syriaca* (Brunner von Wattenwyl, 1861) and *Heteracris morbosa* (Serville, 1838)) families.

We report an information on chromosome sets and C-banding patterns of studied species. *P. syriacus* and *T. pulchripennis* have karyotypes consisted of 19 acrocentric chromosomes in male ($2n\sigma = 18+X0$; NF = 19). Karyotypes of *H. syriaca* and *H. morbosa* consisted of 23 acrocentric chromosomes in mail ($2n\sigma = 22+X0$; NF = 23). All studied species have $X0\sigma/XX\sigma$ sex determination. Analysis of the localization and size of C-positive regions revealed only pericentromeric C-blocs in chromosomes of studied species.

Резюме. Впервые изучены цитогенетические особенности некоторых видов саранчовых из Израиля. Мы провели сравнительный цитогенетический анализ четырёх видов саранчовых, два из которых (*Tmethis pulchripennis* (Serville, 1838) и *Prionosthenus syriacus* (Brisout de Barneville, 1854)) принадлежат к семейству Pamphagidae, а ещё два (*Heteracris syriaca* (Brunner von Wattenwyl, 1861) и *Heteracris morbosa* (Serville, 1838)) — к семейству Acrididae.

В работе представлена информация о хромосомных наборах и локализации С-гетерохроматина у изученных видов. *P. syriacus* и *T. pulchripennis* имеют кариотипы, состоящие из 19 акроцентрических хромосом ($2n\sigma = 18 + X0$; NF = 19). Кариотипы *H. syriaca* и *H. morbosa* состоят из 23 акроцентрических хромосом ($2n\sigma = 22 + X0$; NF = 23). Все изученные виды имеют хромосомное определение пола $X0\sigma/XX\sigma$. Анализ локализации и размера С-позитивных районов хромосом выявил только прицентромерный гетерохроматин. При этом у исследованных представителей семейства Pamphagidae С-блоки — очень мелкие, точечные, а у Acrididae явственные, хорошо идентифицируемые на всех стадиях мейоза.

Introduction

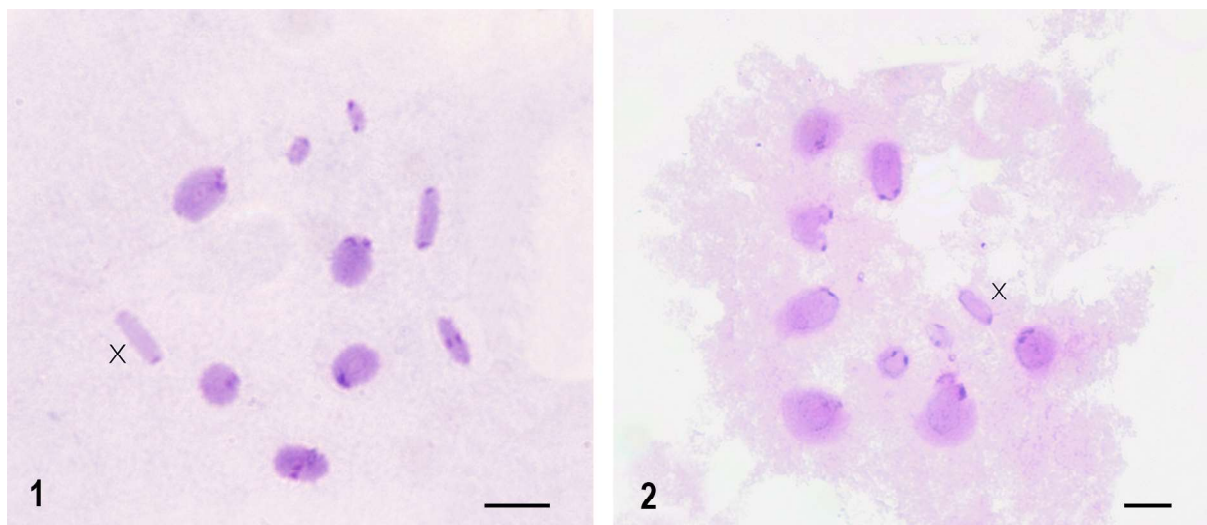
To date, the karyotypic features of Israel grasshoppers remain extremely poorly studied. And this is while grasshoppers have long been the most favorite object of cytogenetic studies. These studies have demonstrated that comparative cytological data on a wide range of taxa, even on such basic parameters as the number and morphology of chromosomes, can provide important information on the systematics, evolution and phylogeny of these insects [White, 1973; Hewitt, 1979; Husemann et al., 2022]. Only *Calliptamus palaestinensis* Ramme, 1930 from Israel has been karyotyped in detail due to the revealed the additional (B-) chromosomes in its set [Nur, 1963].

This article presents new data on the karyotypes of some Israel grasshoppers. We hope that this study will serve as motivation for cytogenetic studies of Israel grasshoppers.

Materials and methods

Material collection. Seven males of *Tmethis pulchripennis* (Serville, 1838) were collected in the spring season (23 to 27 March, 2023) in Negev desert in different loci near Be'er Sheva city. One male of *Prionosthenus syriacus* (Brisout de Barneville, 1854) and one male of *Heteracris syriaca* (Brunner von Wattenwyl, 1861) were collected 22 March, 2023 in surrounded of Kiryat Gat city. One male of *Heteracris morbosa* (Serville, 1838), was caught in the vicinity of Eilat, 19 March, 2023.

Fixation and C-banding. The 0.1 % colchicine solution was injected into abdomens of collected males. After 1.5–2.0 hours, their testes were dissected and placed into



Figs 1–2. C-banded metaphase I of the meiosis of the grasshoppers. 1 — *Prionosthenus syriacus*, 2 — *Tmethis pulchripennis*. Scale bar: 10 μ m.

Рис. 1–2. С-дифференциальное окрашивание хромосом на стадии метафазы мейоза I саранчовых. 1 — *Prionosthenus syriacus*, 2 — *Tmethis pulchripennis*. Масштаб: 10 мкм.

0.9 % solution of sodium citrate for 20 minutes. Then testes were fixed in 3:1 ethanol:glacial acetic acid for 15 minutes. Thereafter, fixed testes were stored in 70 % ethanol in a refrigerator at 4 °C until studied. Air-dried chromosome preparations were made by squashing testis follicles in 45 % acetic acid and then freezing them in dry ice.

The heterochromatin blocks were identified by C-banding, using the technique described by Sumner [1972] with minor modifications. Slides were treated with 0.2 N HCL for 15–20 min, then incubated in the saturated solution of Ba(OH)₂ at 61 °C for 3–5 min, rinsed in tap water and incubated in 2×SSC at 61 °C for 60 min. After being rinsed in distilled water the slides were stained with 2 % Giemsa solution.

Results

The chromosome sets of *Tmethis pulchripennis* (Serville, 1838) and *Prionosthenus syriacus* (Brisout de Barneville, 1854) consist of 9 pair of acrocentric autosomes and one unpaired acrocentric X chromosome in males ($2n^{\sigma} = 19$; 18AA + X) (Fig. 1). The karyotype structure is represented by five large (L_1 – L_5) two medium (M_6 – M_7) and two small (S_8 – S_9) pair of autosomes and the medium size X chromosome (Figs 1–2). At meiotic prophase in *T. pulchripennis* and *P. syriacus* each large bivalents usually form two, rarely one chiasma, medium and small bivalents form one chiasma (Figs 1–2).

Very small pericentromeric C-heterochromatic blocks in all chromosomes of the set were revealed (Figs 1–2).

The karyotypes of *Heteracris syriaca* (Brunner von Wattenwyl, 1861) and *Heteracris morbosa* (Serville, 1838) consist of 11 pairs of acrocentric autosomes and an unpaired acrocentric X chromosome ($2n = 22 + X0^{\sigma}/XX^{\sigma}$). Four autosome pairs are large (L_1 – L_4), five medium autosome pairs (M_5 – M_9) and two autosome pairs

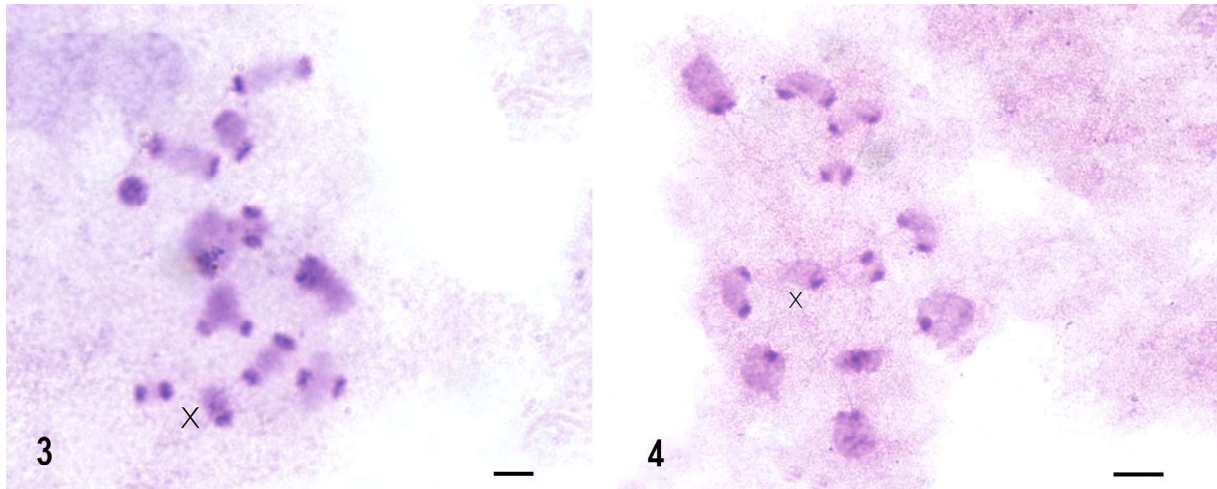
are small sized (S_{10} , S_{11}). The X chromosome is medium sized (Figs 3–4). At meiotic prophase in *H. syriaca* and *H. morbosa* each large bivalents usually form two, rarely one chiasma, medium and small bivalents form one chiasma (Figs 3–4).

C-banding revealed distinct pericentromeric C-blocks in all of the autosome bivalents and the X chromosome (Figs 3–4). S_{11} chromosome in *H. syriaca* is completely heterochromatic.

Discussion

Our cytogenetic study of previously unstudied grasshoppers from Israel confirmed that species belonging to Pamphagidae ($2n^{\sigma}=19$, FN=19) and Acrididae ($2n^{\sigma} = 23$, FN = 23) differ discretely in the number of chromosomes in the karyotype. The results obtained neither support nor refute the previously proposed hypotheses of the evolution of the grasshopper's karyotype from the predominant ($2n^{\sigma} = 23$) in the superfamily Acridoidea to the derivative ($2n^{\sigma} = 19$) in the family Pamphagidae [White, 1973; Hewitt, 1979]. At the same time, it should be emphasized that the chromosome sets of *Tmethis pulchripennis* (Serville, 1838) and *Prionosthenus syriacus* (Brisout de Barneville, 1854) from the fauna of Israel have the plesiomorphic type of chromosome set for the Pamphagidae family ($2n^{\sigma} = 19$), while in the fauna of the Eastern Mediterranean revealed the derivative variants of karyotypes as a result of various types of chromosomal rearrangements [Bugrov, Grozeva, 1998; Bugrov et al., 2016; Jetybayev et al., 2017; Buleu et al., 2020].

As for the studied species of the Acrididae family from the subfamily Eypreponeminae (*H. syriaca* and *H. morbosa*), they have typical features of the chromosome sets described for other species of this taxonomic group [Cabrero et al., 2003].



Figs 3–4. C-banded metaphase I of the meiosis of the grasshoppers. 3 — *Heteracris syriaca*, 4 — *Heteracris morbosa*. Scale bar: 10 μ m.

Рис. 3–4. С-дифференциальное окрашивание метафазных хромосом на стадии метафазы мейоза I саранчовых. 3 — *Heteracris syriaca*, 4 — *Heteracris morbosa*. Масштаб: 10 мкм.

Acknowledgements

The authors are grateful to Israel volunteer Vera Orenov for grasshoppers collecting and Dr. Mazin Qumsiyeh, Bethlehem University for taxonomic identification of *Heteracris morbosa* (Serville, 1838).

This work was carried out under the Federal Fundamental Scientific Research Program (grant No. 1021051703269-9-1.6.12) via the Institute of Systematics and Ecology of Animals SB RAS.

References

- Bugrov A., Grozeva S. 1998. Neo-XY chromosome sex determination in four species of the pamphagid grasshoppers (Orthoptera, Acridoidea, Pamphagidae) from Bulgaria // *Caryologia*. Vol.51. No.2. P.115–121. <https://doi.org/10.1080/00087114.1998.10589126>.
- Bugrov A.G., Jetybayev I.E., Karagyan G.H., Rubtsov N.B. 2016. Sex chromosome diversity in Armenian toad grasshoppers (Orthoptera, Acridoidea, Pamphagidae) // *Comparative Cytogenetics*. Vol.10. P.45–59. <https://doi.org/10.3897/CompCytogen.v10i1.6407>.
- Buleu O.G., Jetybayev I.Y., Mohsen M.-N., Bugrov A.G. 2020. Karyotypes diversity in some Iranian Pamphagidae grasshoppers (Orthoptera, Acridoidea, Pamphagidae): new insights on the evolution of the neo-XY sex chromosomes // *Comparative Cytogenetics*. Vol.14. No.4. P. 549–566. <https://doi.org/10.3897/compcytogen.v14.i4.53688>.
- Cabrero J., Bugrov A., Warchalowska-Sliwa E., Lypez-Leyn M.D., Perfectti F., Camacho J.P.M. 2003. Comparative FISH analysis in five species of Eypreocnemidinae grasshoppers // *Heredity*. Vol.90. P.377–381.
- Hewitt G.M. 1979. Grasshoppers and cricket // John B. (Ed.): *Animal cytogenetics*, 3. Insecta I. Orthoptera. Borntraeger, Berlin, Stuttgart. P.1–170. <https://doi.org/10.3897/compcytogen.v5i4.2307>.
- Husemann M., Lara-Sophie D., David S., Ueshima N., Hawlitschek O., Song H., Weissman D.B. 2022. Evolution of chromosome number in grasshoppers (Orthoptera: Caelifera: Acrididae) // *Organisms Diversity & Evolution*. Vol.22. P.649–657. (2022) <https://doi.org/10.1007/s13127-022-00543-1>.
- Jetybayev I.E., Bugrov A.G., Ünal M., Buleu O.G., Rubtsov N.B. 2017. Molecular cytogenetic analysis reveals the existence of two independent neo-XY sex chromosome systems in Anatolian Pamphagidae grasshoppers // *BMC Evolutionary Biology*. Vol.17. No.1. P. 20. <https://doi.org/10.1186/s12862-016-0868-9>.
- Nur U. 1963. A Mitotically unstable supernumerary chromosome with an accumulations mechanisms in a grasshoppers // *Chromosoma (Berl.)*. Vol.14. P.407–422.
- Sumner A.T. 1972. A simple technique for demonstrating centromeric heterochromatin // *Experimental Cell Research*. Vol.75. P.304–306.
- White M.J.D. 1973. *Animal Cytology and Evolution*. Cambridge. 961 p.

Поступила в редакцию 3.6.2024