

# Cytotaxonomy of the Maculata subcomplex (Hemiptera, Triatominae)

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(With 1 figure)

## 1. Scientific Note

Triatomines are hematophagous insects belonging to the Hemiptera order and Triatominae subfamily. All members of this subfamily are potential vectors of Chagas disease (Tartarotti et al., 2006). The Maculata subcomplex is composed of four species: *Triatoma maculata*, *T. pseudomaculata*, *T. arthurneivai* and *T. wygodzinskyi*. Recent phylogenetic analyzes showed conflicting results in relation to monophyly of this complex, because Justi et al. (2014) have failed to rescue the monophyly of the subcomplex and observed that *T. maculata* was more related to the species of the Brasiliensis subcomplex than the species of Maculata subcomplex (*T. arthurneivai* was not used in the study).

By means of cytogenetic characterization of the spermatocytes (prophase I), Santos et al. (2007) proposed that *T. maculata*, *T. pseudomaculata* and *T. arthurneivai* presented different heterochromatin patterns distribution and meiotic behavior. Panzera et al. (2010) describe the pattern heterochromatic of *T. wygodzinskyi* as identical to that observed for *T. arthurneivai*. However, Carbajal-de-La-Fuente et al. (2010) highlight that in many works of literature *T. wygodzinskyi* was erroneously classified as *T. arthurneivai*. Thus, this work describes the heterochromatic pattern of all species of the Maculata subcomplex, with cytotaxonomic focus.

At least two males from each species were used. They had been assigned by the FCFAR/UNESP, Araraquara, São Paulo, Brazil, FIOCRUZ, Rio de Janeiro, Brazil and CPqRR-FIOCRUZ, Minas Gerais, Brazil. The seminiferous tubules of adult males were torn and fixed to a cover slip. They then underwent the cytogenetic technique of C-banding (Sumner, 1972) and analyzed using a Jenaval light microscope (Zeiss).

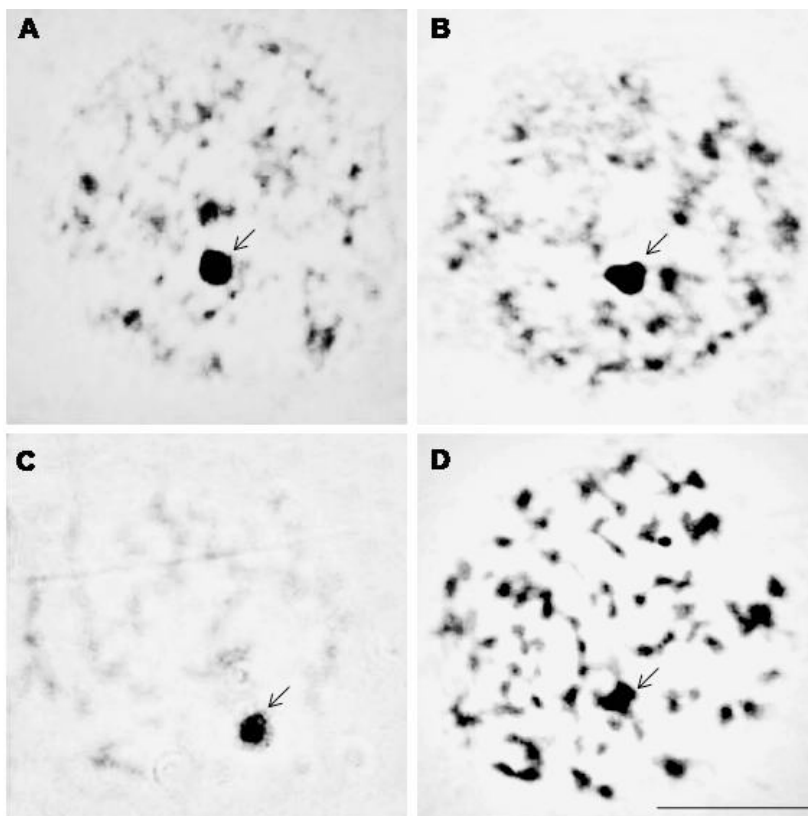
By analyzing of the distribution of the pattern of constitutive heterochromatin in spermatocytes in prophase I has been possible to differentiate all species of Maculata subcomplex: *T. maculata* (Figure 1A) showed a heterochromatic chromocenter formed by the sex chromosomes (arrow) and small heterochromatic blocks dispersed in nucleus;

*T. pseudomaculata* (Figure 1B) showed a heterochromatic chromocenter formed by the sex chromosomes plus one pair of autosomes (arrow) and some heterochromatic blocks dispersed in the nucleus; *T. wygodzinskyi* (Figure 1C) showed only one heterochromatic chromocenter formed by the sex chromosomes (arrow) and *T. arthurneivai* (Figure 1D) showed a heterochromatic chromocenter formed by the sex chromosomes (arrow) and large heterochromatic blocks dispersed in the nucleus.

Schofield (1988) suggests that *T. maculata* and *T. pseudomaculata* species are derived from a common ancestor that was dispersed passively through the feathers of migratory birds and isolated geographically. These species are very similar from the morphological point of view (Corrêa and Espinola, 1964) and cytogenetic analysis described above confirm the data initially described by Santos et al. (2007) and corroborate the specific status of these triatomines. We suggest that during the speciation of *T. pseudomaculata*, heterochromatin loss events were involved in genomic reorganization with loss of heterochromatin, as has been observed for the pallescens group (Alevi et al., 2015).

The phylogenetic relationship between *T. maculata* and Brasiliensis subcomplex reported by Justi et al. (2014) can be confirmed by standard heterochromatin of the species. However, the meiotic behavior of these vectors is different, since during prophase *T. maculata*, the chromocenter is formed by sex chromosomes, different of the species the Brasiliensis subcomplex which has the chromocenter form by sex chromosomes plus one pair of autosomes.

*T. wygodzinskyi* was described based on specimens collected in Santa Rita de Caldas, in southern Minas Gerais, Brazil and initially classified as *T. arthurneivai* (Lent, 1951). Different from the cytogenetic characteristics described initially, our results show that these species display large differences in heterochromatic pattern being the genetic material of *T. arthurneivai* rich in constitutive heterochromatin. Thus, we emphasize that the specimens



**Figure 1.** Constitutive heterochromatin in spermatocytes (prophase I) of all species of Maculata subcomplex. (A) Note the heterochromatic chromocenter formed by the sex chromosomes (arrow) and small heterochromatic blocks dispersed in *T. maculata*; (B) Note the heterochromatic chromocenter formed by the sex chromosomes plus one pair of autosomes (arrow) and some heterochromatic blocks dispersed in *T. pseudomaculata*; (C) Note with only one heterochromatic chromocenter formed by the sex chromosomes (arrow) in *T. wygodzinskyi*; (D) Note the heterochromatic chromocenter formed by the sex chromosomes (arrow) and large heterochromatic blocks dispersed in *T. arthurneivai*. Bar: 10  $\mu$ m.

analyzed by Santos et al. (2007) were *T. wygodzinskyi* and not *T. arthurneivai* as classified by the authors.

Thus, in the present study demonstrated that the analysis of heterochromatic pattern is a cytotaxonomic tool of utmost importance to characterize the species of Maculata subcomplex.

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