

## Reproductive aspects of Chagas disease vectors (Hemiptera, Triatominae) with anatomical teratologies



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### ARTICLE INFO

#### Keywords:

Spermatogenesis  
Gonadal dysgenesis  
Morphological abnormalities  
Triatomines

### ABSTRACT

As Chagas disease has no cure, vector control is the most effective method of preventing this neglected disease. Generally the anatomical teratologies are associated with hormonal dysfunction of the *corpus allatum* (juvenile hormone), presents genetic basis or unfavorable conditions, and are recessive and not sex linked. Thus, we characterize the male gonads and spermatogenesis of triatomines with anatomical teratologies to evaluate if the malformations interfere in the gametogenesis of these vectors. All teratogenic triatomines analyzed did not present presented gonadal dysgenesis and alterations in spermatogenesis. Thus, we characterize the presence of anatomical teratologies in some species of triatomines and demonstrate that these malformations in the external morphology do not interfere in the gonads and gametogenesis of these vectors. In addition, although new studies with the *corpus allatum* should be conducted we suggesting that the use of juvenile hormones does not present effectiveness in the reproductive control of these vectors.

Chagas disease is a neglected disease distributed in endemic areas of 21 Latin American countries caused by the protozoan *Trypanosoma cruzi* and transmitted to humans mainly by contact with feces of triatomines (WHO, 2015). Currently, there are 152 species of triatomines, distributed in 18 genera and five tribes, being all species considered as potential vector of Chagas disease (Justi and Galvão, 2017; Oliveira and Alevi, 2017).

As Chagas disease has no cure and treatment is effective only in the acute phase of the disease (which is often asymptomatic), vector control is the most effective method of preventing this neglected disease (WHO, 2015). Thus, all knowledge about these hematophagous insects is important and can generate subsidies to assist the vector control programs.

Anatomical studies of the triatomines are common and have contributed to the taxonomic, systematic and evolutionary knowledge of the vectors (Pinto, 1927; Galvão, 2014). However, little is known about anatomical teratologies in the subfamily Triatominae, being the majority of studies performed in *Triatoma infestans* (Carcavallo, 1967; Juárez, 1972; Carcavallo and Galíndez Girón, 1995; Jurberg et al.,

1997). The teratologies are present in the structures of the head, thorax and abdomen of triatomines (Carcavallo et al., 1998).

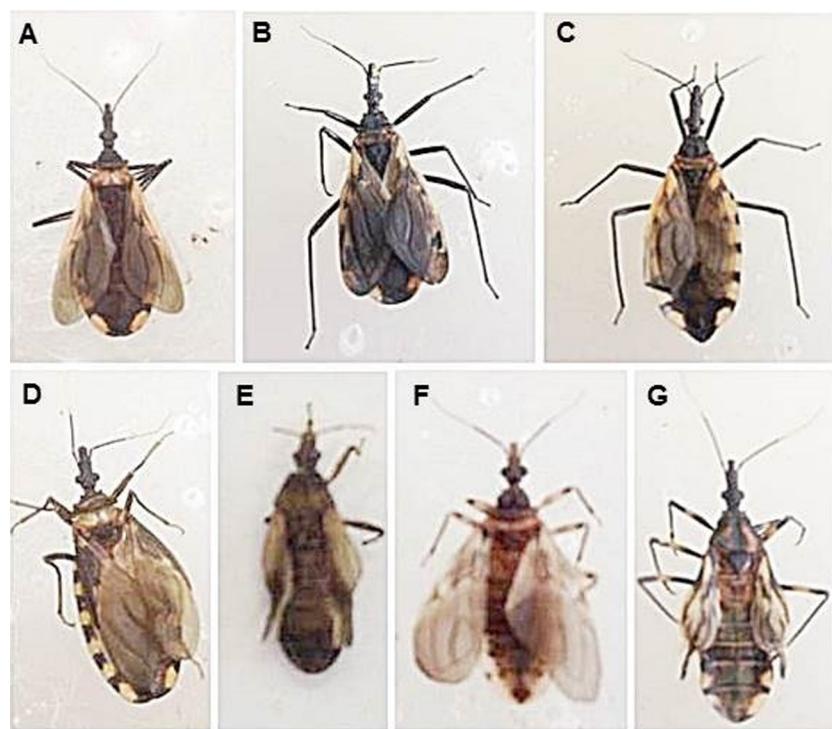
Although the teratologies of the triatomines generally are associated with hormonal dysfunction of the *corpus allatum* (responsible for producing juvenile hormone) (Juárez, 1972), the causes of their occurrence are very diverse (genetic basis or unfavorable conditions of the breeding environment), as described by Ryckman (1971). Triatomines exposed to 9-cis-retinoic acid (9cisRA) or juvenile hormone underwent profound morphological changes upon moult, generating abnormal 5th instar nymphs and also markedly increased the death of insects during the moulting (Nakamura et al., 2007). Ryckman (1971) performed experimental crosses between teratogenic *T. protracta* and described the teratologies as recessive and not sex linked.

Thus, in order to contribute to the knowledge of these insects of importance to public health, we characterize the male gonads and spermatogenesis of triatomines with anatomical teratologies to evaluate if the malformations interfere in the gametogenesis of these vectors.

Were analyzed at least two adult male specimens of each species (*T. costalimai*, *T. garciabesi*, *T. guasayana*, *T. infestans*, *T. juazeirensis*, *T.*

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**Fig. 1.** Examples of anatomical teratologies in triatomines: (A) *T. klugi*, (B) *T. rubrovaria*, (C) *T. pseudomaculata*, (D) *T. infestans*, (E) *P. tertius*, (F) *T. guasayana* and (G) *T. brasiliensis*. Note the presence of teratology in the wings of all species (A–G), as well as in the thorax of *T. klugi* (A) and *T. pseudomaculata* (D) and in the abdomen of *P. tertius* (E) and *T. brasiliensis* (G).

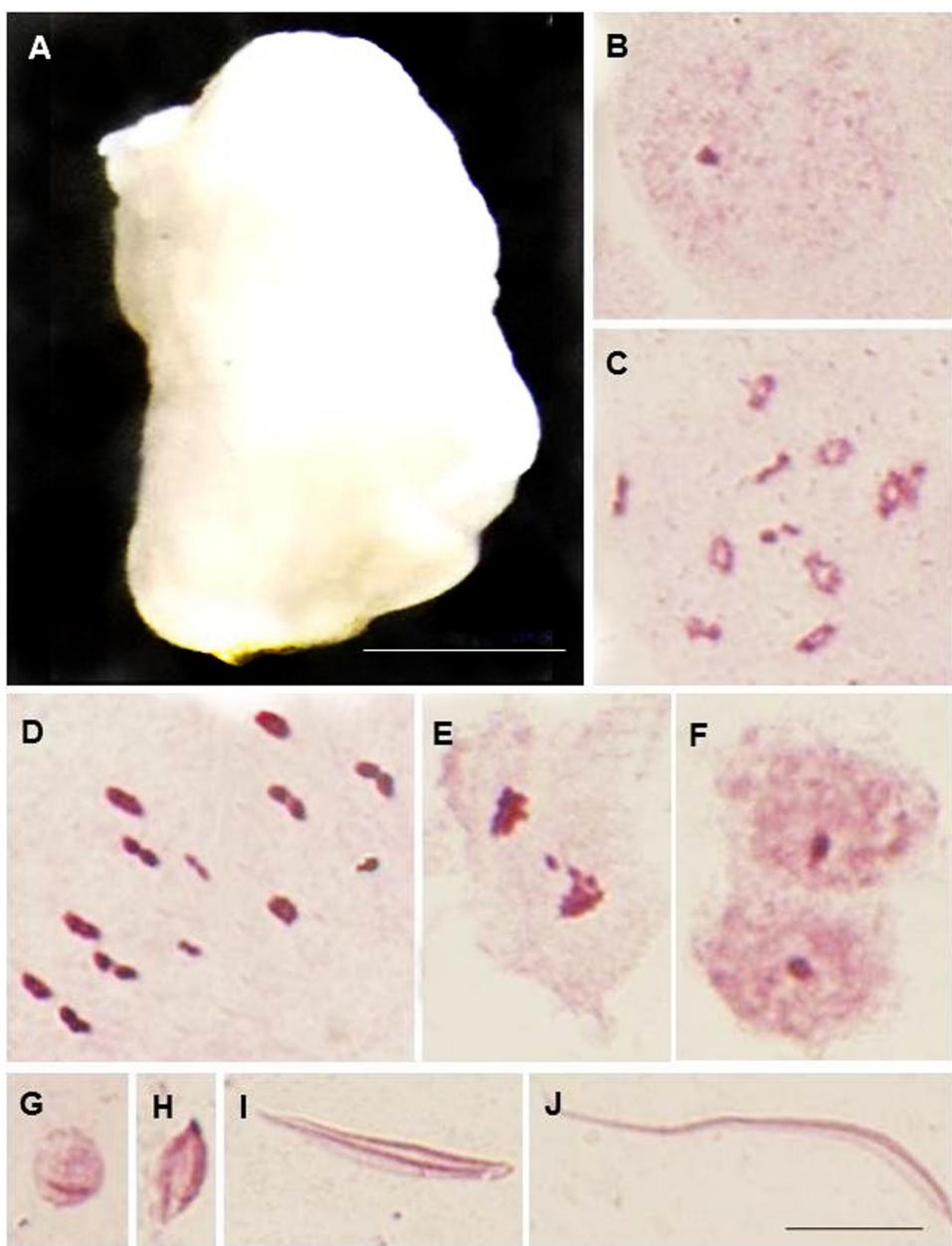
**Table 1**  
Teratologies described in triatomines.

Triatomines	Teratologies	References
<i>Triatoma brasiliensis</i>	abdomen, wings and legs (tibia)	Carcavallo et al. (1998)
<i>Triatoma breyeri</i>	thorax (pronotum)	Carcavallo et al. (1998)
<i>Triatoma costalimai</i>	wings	First description
<i>Triatoma garciabesi</i>	wings	First description
<i>Triatoma guasayana</i>	wings	First description
<i>Triatoma infestans</i>	wings (corium), legs (tibiae and tarsi), head (ocellar and antenna), thorax (pronotum, mesonotum, metanotum and propleura), adultoids (treatment of anti-juvenile hormonal compound)	Carcavallo (1967); Juárez (1972), Carcavallo and Galíndez Girón (1995), Jurberg et al. (1997)
<i>Triatoma juazeirensis</i>	legs and wings	First description
<i>Triatoma klugi</i>	wings and thorax	First description
<i>Triatoma lecticularia</i>	legs and thorax	First description
<i>Triatoma lenti</i>	legs and wings	First description
<i>Triatoma maculata</i>	abdomen and wings	Carcavallo et al. (1998)
<i>Triatoma patagonica</i>	thorax (mesonotum and metanotum) and wings	Carcavallo et al. (1998)
<i>Triatoma protracta</i>	thorax (pronotum and mesonotum), wings and genital	Ryckman (1971)
<i>Triatoma pseudomaculata</i>	wings	First description
<i>Triatoma rubida</i>	legs (an oligometry with unilateral atrophy in the left mesothoracic)	Faúndez and Ayala (2017)
<i>Triatoma rubrovaria</i>	wings	First description
<i>Triatoma sordida</i>	wings	Carcavallo et al. (1998)
<i>Triatoma vitticeps</i>	legs and wings	First description
<i>Panstrongylus megistus</i>	adultoids (treatment of anti-juvenile hormonal compound)	Jurberg et al. (1982)
<i>Psammolestes coreodes</i>	legs and thorax	First description
<i>Psammolestes tertius</i>	abdomen and wings	First description
<i>Panstrongylus humeralis</i>	legs (a unilateral atrophy on the right metathoracic)	Faúndez and Ayala (2017)
<i>Rhodnius brethesi</i>	legs and wings	First description
<i>Rhodnius ecuadoriensis</i>	wings	Carcavallo et al. (1998)
<i>Rhodnius neglectus</i>	legs	First description
<i>Rhodnius pictipes</i>	legs	First description
<i>Rhodnius prolixus</i>	legs and wings	First description
<i>Rhodnius robustus</i>	adultoids (treatment of anti-juvenile hormonal compound)	Jurberg et al. (1997)
	legs (femur and tibia) and wings corium	Carcavallo et al. (1998)

*klugi*, *T. lecticularia*, *T. lenti*, *T. pseudomaculata*, *T. rubrovaria*, *T. vitticeps*, *Psammolestes coreodes*, *P. tertius*, *Rhodnius brethesi*, *R. neglectus*, *R. pictipes* and *R. prolixus*) provided by the “Insetário de Triatominae”, from FCFAR/UNESP, Araraquara, São Paulo, Brazil. The selection of triatomines occurred based on the presence of anatomical teratologies. The insects were dissected, the testicles were removed, and examined by

stereoscope microscope for analysis of gonadal dysgenesis (unilateral or bilateral dysgenesis) (Almeida and Carareto, 2002). Posteriorly, the seminiferous tubules were shredded, smashed, set on a slide in liquid nitrogen and stained with lactic acetic orcein (De Vaio et al., 1985; Alevi et al., 2012) for analysis of spermatogenesis.

Some species were selected to represent the anatomical teratologies



**Fig. 2.** Testis and spermatogenesis of *T. rubrovaria*. (A) Normal testis. Bar: 10 mm. (B, C) Prophase. (D) Metaphase. (E) Anaphase. (F) Telophase. (G–J) Spermatogenesis. Bar: 10  $\mu$ m.

(Fig. 1). All triatomines analyzed presented teratologies in the wings (Table 1, Fig. 1A–G). In addition, malformations in the thorax (Table 1, Fig. 1A, D) and abdomen (Table 1, Fig. 1E, G) were also observed. On the other hand, none of these triatomines presented gonadal dysgenesis and alterations in spermatogenesis [represented by the gonad (Fig. 2A) and spermatogenesis (Fig. 2B–J) of *T. rubrovaria*].

These results demonstrate that triatomines with anatomical teratologies do not present gonadal dysgenesis and alterations in gametogenesis. Although the fertility reduction may be related to other factors, such as inability to transfer sperm to the female's spermathecal (Perlowagora-Szumlewicz and Correia, 1972), or the existence of essential secretions for sperm migration to the female receptacle (Schreiber et al., 1975), we demonstrated for the first time that the formation of gametes in teratogenic occurs normally, corroborating the results of Ryckman (1971).

The use of products analogous to juvenile hormones was initially associated with the control of triatomines in domiciliary regions

(Pinchin et al., 1978; Carcavallo et al., 1998). Several studies with these analogues have demonstrated that the application leads to anatomical teratologies and the formation of adultoids (Table 1). Considering the hypothesis that the anatomical changes come only from problems in the *corpus allatum* and consequently in the rates of juvenile hormone, the crosses performed by Ryckman (1971) in conjunction with the reproductive aspects described here demonstrated that males triatomines with anatomical teratologies present reproductive fitness, suggesting that the use of juvenile hormones does not present effectiveness in the reproductive control of these vectors. However, further studies should be performed with this structure to determine whether triatomine teratogens are exclusively associated with hormonal dysfunction or if there are other influences, as already mentioned above.

Thus, we characterize the presence of anatomical teratologies in some species of triatomines and demonstrate that these malformations in the external morphology do not interfere in the gonads and gametogenesis of these vectors. In addition, although new studies with the

*corpus allatum* should be conducted (to confirm its direct relationship with the teratologies), suggesting that the use of juvenile hormones does not present effectiveness in the reproductive control of these vectors.

### Ethical standards

The experiments comply with the current laws of the country in which they were performed.

### Conflict of interest

The authors declare that they have no conflict of interest.

### Acknowledgments

The study was supported by Fundação de Amparo à Pesquisa do Estado de São Paulo (Process numbers 2013/19764-0) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, Brazil).

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