




A case of gynandromorphism in *Rhipicephalus sanguineus* s.l. from Mexico

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Abstract

We report the presence of a brown dog tick (*Rhipicephalus sanguineus* s.l.) gynandromorph collected inside a house in the state of Guanajuato, Mexico. This work provides the first report of gynandromorphism in a tick from Mexico, and represents the third report of this condition in *R. sanguineus* s.l. in the world.

Keywords Gynandromorph · Brown dog tick · Morphological abnormality · Tropical lineage

Introduction

Gynandromorphism is a rare process seen in arthropods where a specimen exhibits both male and female characters simultaneously; this is considered to be an abnormal process of embryonic development, with the unequal distribution of sex-linked chromosomes, particularly in mandibulates (insects) and chelicerates (spiders and ticks) (Homsher and Yunker 1981; Pereira et al. 2010). In the case of ticks, this phenomenon has been documented extensively around the world, with approximately 80 documented cases, mainly in hard ticks (Guglielmone et al. 1999; Labruna et al. 2000, 2002; Keskin et al. 2012; Muñoz-Leal et al. 2018).

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The traditional classification of Campana-Rouget (1959) considers the existence of five forms of gynandromorphism in ticks: (1) bipartite protogynander, where the external characters of male and female ticks are equally represented; (2) deutergynander, where characters of one sex are decreased to a quadrant; (3) metagynander, where characters of one sex are decreased to a small segment; (4) gynander intriqué, a protogynander or deutergynander in which some pieces of male or female characters are embedded in areas of the opposite sex; and (5) mosaic gynandromorphism, where there is no definitive line separating one sex from the other, but the pieces are entangled without any indication of symmetry. It has been recognised that the bipartite protogynander is the most widely documented phenomenon (Labruna et al. 2002; Keskin et al. 2012).

Gynandromorphism has been recorded in eight taxa of the *Rhipicephalus* genus (*R. annulatus*, *R. appendiculatus*, *R. bursa*, *R. geigy*, *R. kochi*, *R. microplus*, *R. simus* [probably *R. praetextatus*], and *R. sanguineus* s.l.) in seven countries of the Afrotropical (Mozambique, South Africa), Neotropical (Argentina, Brazil, Guatemala), and Palearctic (Portugal, Russia) regions (Pereira and Castro 1945; Campana-Rouget 1959; Gothe 1967; Mnase et al. 1987; Guglielmone et al. 1999; Labruna et al. 2002). Here, we describe the first case of a gynandromorphic *R. sanguineus* s.l. in Mexico.

Materials and methods

As a part of the ‘National Program for *Rickettsia* Surveillance’, we received brown dog ticks (*R. sanguineus* s.l.) from the National Network of Public Health Laboratories of the state of Guanajuato. Ticks were collected by the vectors brigade on 18 July 2018, inside houses with the presence of dogs in the locality of Paso de Guillermo, Municipality of Xichú, in the state of Guanajuato (99°55'30"N, 21°13'17"W, 1380 m above sea level). Ticks were collected manually and fixed in 70% ethanol until processing in the laboratory. Morphological identification of ticks was performed with an Olympus SZX7 stereoscopic microscope using specialised taxonomic keys (Keirans and Litwak 1989; Dirección General de Salud Animal 1992).

Because *R. sanguineus* encompasses a complex of species and the existence of temperate and tropical lineages on the American continent has been described, it is essential to implement molecular tools to discriminate them (Nava et al. 2018). To identify the lineage to which the tick corresponded, DNA extraction was performed from a single leg which was crushed using a sterile pestle. Genomic DNA was extracted using the QIAamp® DNA Mini Kit (Qiagen, Hilden, Germany). Molecular identification of the tick was performed through the amplification of a partial DNA sequence (≈ 440 bp) of the mitochondrial 16S rDNA gene, using the protocols of Norris et al. (1996). The reaction mixture consisted of 12.5 μ l GoTaq® Green Master Mix 2X (Promega, Madison, WI, USA), 100 ng of each primer, 6.5 μ l nuclease-free water and 200 ng DNA in a final volume of 25 μ l (Sánchez-Montes et al. 2016). PCR products were resolved on 2% agarose gels stained with Smart-glow (Accuris Instruments, cat. E4500-PS) and visualised using an Accuris Smartdoc photodocumenter. The purified amplification product was submitted for sequencing at the Pathogen Genome Laboratory of the Instituto de Diagnóstico y Referencia Epidemiológicos in an Applied Biosystems 3500 Series sequencer. The sequence was compared to those reported in GenBank using the Basic Local Alignment Search Tool (BLAST).

The sequence obtained in this study was deposited in GenBank with accession number MT322611.

Results and discussion

We received 11 *R. sanguineus* s.l., 10 normal female specimens, and one gynandromorph identified. We identified the presence of a deutero gynander *R. sanguineus* s.l. with the following characteristics:

[Dorsum] Capitulum symmetrically divided, right palp slightly larger with equally sized porous areas (Fig. 1a, b). Scutum with the typical punctuations of this species, right margin under the right eye subtly irregular (Fig. 1b). Presence of four festoons and comma-shaped spiracular plate in the left dorsal quadrant of the male, below the fourth pair of legs (Fig. 1c–e).

[Venter] Genital and anal apertures presented by both sexes, poorly developed in female (Fig. 1f); male quadrant with a single ad-anal plate (Fig. 1g). Right coxa with internal spur slightly narrower than the left one; broken external spur of the right coxa I. Right coxae II–IV closer together, leaving the coxa IV at the level of the left coxa III (Fig. 1f). Male quadrant below right coxa IV (Fig. 1f).

The sequence generated in this study exhibited a similarity of 99–100% with sequences of the tropical lineage of *R. sanguineus* s.l. available in GenBank, from Mexico (Baja California [KT382472], Chihuahua [MK680295], Oaxaca [MH018821] and Sonora [KT382471]), Colombia (GU553076), Brazil (GU553075) and the USA (KT382476).

To the best of our knowledge, this is the first report of gynandromorphism in a tick from Mexico, and represents the third report of this condition in *R. sanguineus* s.l. in the world (Pereira and Castro 1945; Labruna et al. 2002). On the American continent, the presence of gynandromorphs has been detected in two species of the genus *Rhipicephalus* (*R. microplus* and *R. sanguineus* s.l.) (Guglielmone et al. 1999; Labruna et al. 2002). The two records of gynandromorphs in *R. sanguineus* s.l. have been presented in Brazil and represent a very rare event, given that there was almost 50 years between the reports (Pereira and Castro 1945; Labruna et al. 2002). Additionally, these two records exhibited mosaic and bipartite protogynander, respectively. As far as we know, this record represents the first report of a deutero gynander *R. sanguineus* s.l. worldwide. In other species of the Old World, deutero gynander appears to be the most common gynandromorphism event, as in the case of an *R. simus* (= *praetextatus*) specimen from Mozambique which exhibited the right posterior quadrant with male characters (Campana-Rouget 1959; Gothe 1967), in contrast to the specimen collected in this work. In the only two previous records of gynandromorphs in *R. sanguineus* s.l., the number of ticks collected was not specified, therefore it is not possible to calculate the proportion of normal against gynander specimens in these taxa. Our gynandromorph was one of a sample of 11 ticks (9%), which represents a much higher percentage compared to other records in the Neotropical region, which range from 0.002% (1/42,642) and 0.04 (3/7434) to 1.1% (1/92), in *R. microplus*, *Amblyomma neumanni* and *A. parvitarsum*, respectively, collected in Argentina (Guglielmone et al. 1999; Muñoz-Leal et al. 2018).

It was possible to identify the specimen collected here as corresponding to the tropical lineage of *R. sanguineus* (Nava et al. 2018). Further studies are required to establish the effect of birth defects on tick fitness and ecology.

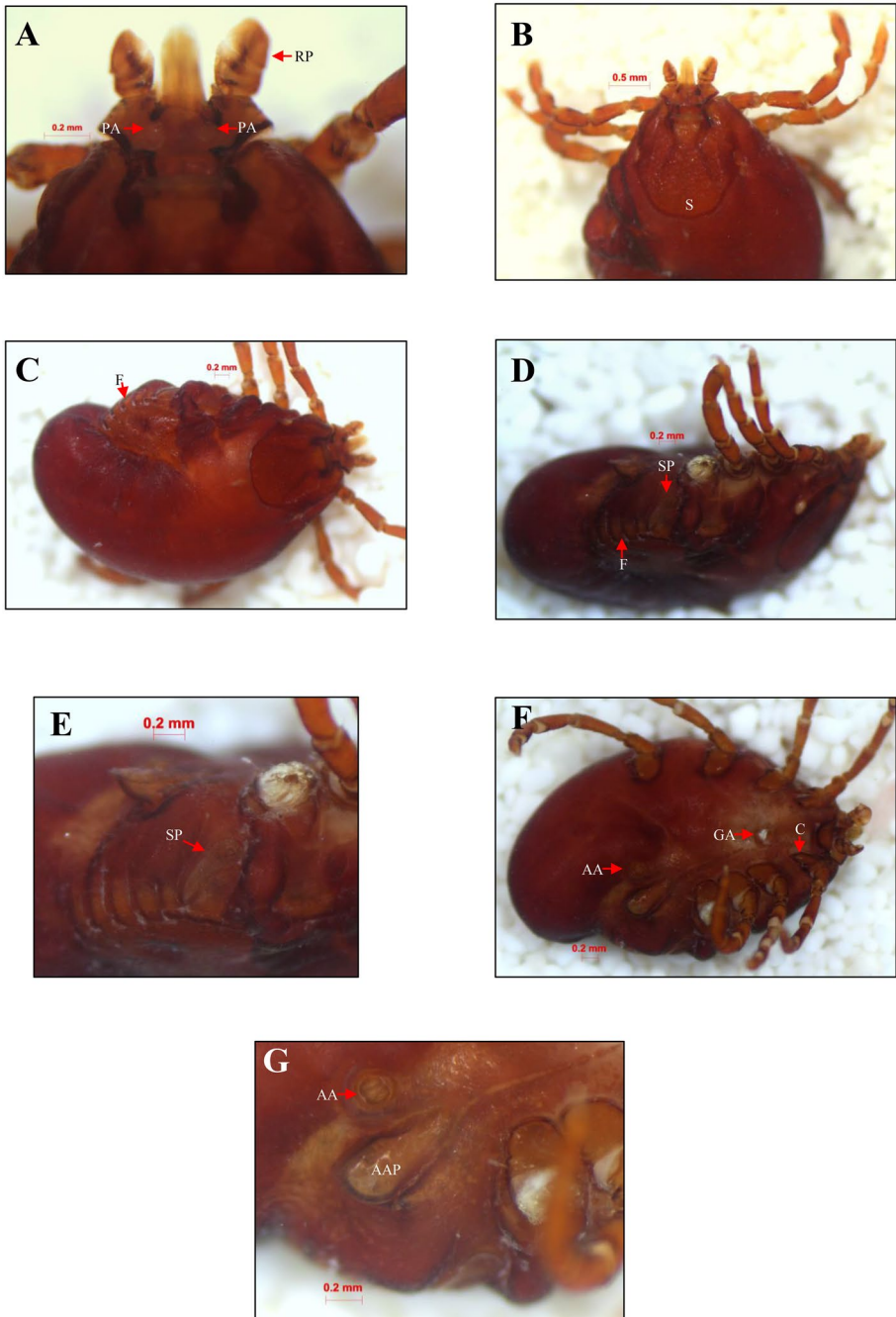


Fig. 1 Morphological characters of *Rhipicephalus sanguineus* s.l. gynandromorph: **a–c** Dorsal view, **d**, **e** Lateral view, **f**, **g** Ventral view. *AA* anal aperture, *AAP* ad-anal plate, *C* coxa, *F* festoons, *GA* genital aperture, *PA* porose area, *RP* right palp, *S* scutum, *SP* comma-shaped spiracular plate

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Data availability Sequences generated in this article are deposited in GenBank.

Compliance with ethical standards

Conflict of interest All the authors declare to have no conflict or competing of interest.

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