



Research Communication

Morphological abnormality in larvae of *Amblyomma oblongoguttatum* (Acari: Ixodidae)

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ABSTRACT

Amblyomma oblongoguttatum is a tick that is a parasite of mammals and its preferred hosts are the wild pigs, tapirs and small rodents, being collected also parasitizing domestic dogs and eventually humans. An *Amblyomma oblongoguttatum* larva with morphological abnormality was collected along with other specimens (larvae and females) of this species and with morphologically normal *Amblyomma naponense* nymphs in a wild pig - *Pecari tacaju* - in a rural area of the state of Rondônia / Brazil, during research of tick-borne disease environments. The morphologically abnormal specimen showed a bifurcation of the posterior region of the opisthosoma, without other altered morphological characters. This is the first record of morphological abnormality at the larval stage of *Amblyomma oblongoguttatum*.

Keywords: Tick, Anomaly, Teratological phenomena, Ixodidae.

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1 Introduction

Morphological changes in ticks are rarely documented events. The main changes described are gynandromorphism, bifurcation and asymmetry in the idiosome, anomalies in the development of the legs, alterations in the oral parts of gnathosoma and asymmetry of peritrematous plaques. Also the appearance, in the same specimen, of two anus, three stigmas or two caudal appendages (NUTTALL, 1914; GOUVEA SOUTO, 1939; NEVEU LEMAIRE, 1938; NOWAK-CHMURA, 2012; KESKIN et al., 2016).

Cases of anomalies arising from natural processes are extremely low in natural environments (ESTRADA-PEÑA, 2001). Among the main interferences that may influence anomalous development are exposure to chemical compounds, temperature and humidity of the air, hosts sensitized with chemicals and problems in the regeneration of tissues after an injury (BUCZEK, 2000; SERRA-FREIRE; BORSOI, 2009; NOWAK-CHMURA, 2012).

Amblyomma oblongoguttatum is a tick that is widely distributed in Brazil, being reported in the states of Amazonas, Rondônia, Acre, Roraima, Pará, Minas Gerais, Goiás, Mato Grosso, Rio de Janeiro and Espírito Santo (ANDREOTTI et al., 2108). It is a parasite of mammals and its preferred hosts are the wild pigs, tapirs and small rodents, being collected also parasitizing domestic dogs and eventually humans in rural areas of the Brazilian Amazon (LABRUNA et al., 2005).

The aim of this report is to present a case of morphological abnormality in an *Amblyomma oblongoguttatum* larvae collected in a wild pig – *Pecari tacaju* – in the rural area of the state of Rondônia, northern region of Brazil.

2 Material and methods

In January 2017, during the surveillance of tick-borne disease environments (OLIVEIRA et al., 2015), 10 (ten) ticks were collected from the ear of a wild pig - *Pecari tajacu* (LINNAEUS, 1758) - captive, in the rural area of the district of Jaci Paraná, city of Porto Velho in the state of Rondônia, Brazil.

The material was sent to the Central Laboratory of Public Health of the State of Rondônia, preserved in 70° alcohol and stereomicroscopically screened, trying to diagnose stage, sex, state of preservation of the specimens and preliminary taxonomic identification. At this moment, it was verified that one of the specimens presented a morphological abnormality and as a flow of diagnostic confirmation is established, proposed by the Ministry of Health of Brazil, these specimens were sent to the National Reference Laboratory in Vectors of the Rickettsioses of the State Oswaldo Cruz Foundation of Rio de Janeiro, for the validation of the morphological identification and molecular analysis of the material.

The taxonomic revision was carried out with the aid of species description (BARBIERI et al., 2012) and adult dichotomous keys (BARROS-BATTESTI et al., 2006), nymphs (MARTINS et al., 2010) and larvae (AMORIM; SERRA-FREIRE, 1999). To aid in the specific identification of larvae (all morphologically equal), the genomic material of one specimen (without morphological anomaly) was extracted by the NaCl extraction technique (ALJANABI; MARTINEZ, 1997), and molecularly identified by the fragment amplification of the second transcribed internal spacer (ITS2). For this, primers were used (MCLAIN et al., 1995; BEATI et al., 2012) and amplification protocol described in the literature (BEATI et al., 2012). The amplicon (~950 pb) was visualized on 2% agarose gel stained with ethidium bromide, the obtained PCR product of the expected size was purified with Kit Wizard® SV Gel and PCR Clean-up System (Promega, Corp., Madison,

WI, U.S.A.) and sequenced on the ABI 3730 automatic DNA analyzer (Applied Biosystems, Inc., Carlsbad, CA, U.S.A.). The sequences obtained were edited and the consensus sequence generated in the program ChromasPro 1.5 (Technelysium PtyLtd., Tewantin, Qld, Australia). The DNA sequence obtained (Access number GenBank MG783327) was identified by similarity evaluation through comparative analysis with the sequences deposited in GenBank with the aid of BLASTN (*Basic Local Alignment Search Tool- Nucleotide*).

Images of the larva with the morphological abnormality were made using the Leica DMC 2900 digital camera coupled to the Leica M205C stereoscopic. This specimen is deposited in the Collection of Apterous Arthropod Vectors of the Importance of Health in Communities - CAVAISC-FIOCRUZ, under number IXO 3327.

3 Results and discussion

The ten specimens collected were identified as two females of *A. oblongoguttatum*, two nymphs of *Amblyomma naponense* and six larvae of *Amblyomma oblongoguttatum*. The ITS2 sequence obtained from the larva showed 99% identity (949/951) with sequence of *A. oblongoguttatum* deposited in GenBank (AY887115) from Rondônia, corroborating the morphological identification. Of these, a larva of *A. oblongoguttatum* was identified with a deformity characterized with sagittal cleft in the opisthosoma, forming a bifurcation that gave it a cordiform aspect (Fig. 1).



Figure 1. Larva of *Amblyomma oblongoguttatum* with morphological abnormality. A. Dorsal view. B. Ventral view.

A malformation in *A. oblongoguttatum* in the northern region of Brazil (State of Pará) has been described by LABRUNA et al., (2000) who verified the gynandromorphism. This is one of the main malformations reported in the genus *Amblyomma* - examples: *Amblyomma latum* by (NOWAK-CHIMURA et al., 2012); *Amblyomma mixtum* by (RIVERA-PAEZ et al., 2015); *Amblyomma dubitatum* by (MARTINS et al., 2017) and *Amblyomma sculptum* (= *Amblyomma cajennense*) by (LABRUNA et al., 2002).

There are also reports of abnormalities for species of Ixodidae of other genera, for example: *Rhipicephalus sanguineus* (LABRUNA et al., 2002; SERRA-FREIRE; BORSOI, 2009), *Rhipicephalus (Boophilus) microplus* (GUGLIELMONE et al., 1999), *Ixodes scapularis* (PRUSINSKI et al., 2015; LARSON; PASKEWITZ, 2016), *Dermacentor andersoni* (DEDRGOUSSOFF; CHILTON, 2007) and for *Haemaphysalis qinghaiesis* (REN et al., 2016).

Despite the bifurcation in the opisthosoma, other structures were not affected in the *A. oblongoguttatum* larva analyzed. The specimen presents

complete gnathosoma, with no duality and this specimen did not show duplication of anal opening.

Considering that the tick with the morphological abnormality was collected in a wild animal from the forests of the region, without occurrence of agriculture and livestock, there are only small rural dwellings the possibility of the malformation observed to be associated with exposure to chemical compounds is low. The wild pig is a natural host of the species (LABRUNA et al., 2005), and it is unlikely that the larva has been fed in a host sensitized with chemicals before being withdrawn from the animal.

4 Conclusion

An *A. oblongoguttatum* larva with posterior bifurcation of the opisthosoma was collected along with other specimens (larvae and females) of this species and with morphologically normal *Amblyomma naponense* nymphs in a wild pig - *Pecari tajacu* - in a rural area of the state of Rondônia, being this the first record of morphological abnormality in the larval stage of *Amblyomma oblongoguttatum* in Brazil.

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