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New records for *Amblyomma rotundatum* Koch (Acari: Ixodidae) from the cane toad, *Rhinella marina* L. in Florida, with notes on identification of the nymphs

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Abstract

Sixty-one cane toads, *R. marina,* were captured by hand at four locations in Dade County, Florida, on May 9 (n=32) and September 3 (n=29) 2014. Toads were examined for ectoparasites and any ticks seen were removed and placed in 95% ethanol. Of the 61 toads, 8 (13.1%) collected in two separate locations were found with attached *Amblyomma* sp. A total of 31 ticks (12 nymphs, 19 larvae) were removed from the toads, ranging from 1-13 specimens collected per toad. In the case of 3 nymphal specimens found in our study, precise identification could not be made by conventional light microscopy due to obscured or unclear characters; therefore, scanning electron microscopy (SEM) was performed on these specimens to aid in identification. SEM analysis revealed an internal spur, albeit tiny, on coxa II of the specimens in question, but no spur was seen on coxa III. Published key characters for *A. rotundatum* state that, "a small internal spur is present on each of coxa II and III." Despite this published description, we concluded that the presence of a small spur on coxa II in this case was diagnostic for *A. rotundatum.*

Key words: Cane Toad; Ticks; Ectoparasites; *Amblyomma rotundatum*

Introduction

Knowledge of ticks and other ectoparasites present in a given area and their hosts is important to parasitologists, biologists, entomologists, and ecologists. Additionally, parasite burdens and vector-borne disease threats from parasites are important factors in wildlife conservation efforts, especially if the hosts are imported or "invasive" (Becklund 1968). The cane toad, *Rhinella marina*, is a large terrestrial toad native to Central and South America that was purposely introduced to interior south-central Florida several times between 1936 and 1944 as a biocontrol agent for beetles in sugar-cane fields (Riemer 1958, Krakauer 1968). It is now well established from the Florida Keys to central Florida (Oliver *et al.* 1993). The tick, *Amblyomma rotundatum* Koch, feeds on a variety of amphibians and reptiles in South America (Brumpt 1924), but in the U.S. it has been found primarily on the cane toad (Oliver *et al.* 1993). This paper provides new records for nymphal *Amblyomma rotundatum* and larval *Amblyomma* spp. (presumably *A. rotundatum*) removed from cane toads collected in the Miami, Florida area.

Materials and methods

Sixty-one *R. marina* were collected at four locations in Dade County, Florida, on May 9 (n=32) and September 3 (n=29) 2014. Collections were made in accordance with approval of the Institutional Animal Care and Use Committee (2013-233, Auburn University), and all individuals were captured by hand, at night, in open green spaces surrounding a suburban area. Toads were examined for ectoparasites, and any ticks seen were removed from the toads by hand, immediately stored in 95% ethanol, and shipped to Mississippi State University (MSU). Specimens were processed at MSU, but ultimately submitted to the last author (RGR) for identification.

Results and discussion

Of the 61 toads, 8 (13.1%) collected in two separate locations were found with attached *Amblyomma* sp. (Table 1). A total of 31 ticks (12 nymphs, 19 larvae) were removed from the toads, ranging from 1-13 specimens collected per toad. For comparison of this infestation, another study showed 3/59 (5.0%) of *R. marina* infested with *A. rotundatum* (Oliver *et al.* 1993). In the case of 3 nymphal specimens in our study (from toads 437 and 283), precise identification could not be made by conventional light microscopy due to obscured or unclear characters described in keys to the nymphal *Amblyomma* species of the United States (Keirans and Durden 1998). Therefore, scanning electron microscopy (SEM) was performed on these specimens to aid in identification. SEM analysis revealed an internal spur, albeit tiny, on coxa II of the specimens in question, but no spur was seen on coxa III (Figure 1). Published key characters for *A. rotundatum* state that, "a small internal spur is present on each of coxa II and III" (Keirans and Durden 1998). Despite this published description, we concluded that the presence of a small spur on coxa II is diagnostic for *A. rotundatum*, since the only other North American *Amblyomma* with which *A. rotundatum* is likely confused is *Amblyomma dissimile* Koch, also a parasite of amphibians and reptiles, but a species lacking internal spurs on both coxa II and coxa III.

TABLE 1. Collection data for ticks removed from cane toads, 2014.

Vial No. (Toad)	Date Collected	Site Collected	GPS Coordinates	Contents
272	May 9	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	7 nymphs *Amblyomma rotundatum*
273	May 9	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	13 larvae *Amblyomma* spp. (most likely *A. rotundatum*)
283	May 9	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	2 nymphs *A. rotundatum*
437	Sep 3	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	2 nymphs *A. rotundatum*
438	Sep 3	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	1 larva *Amblyomma* spp. (most likely *A. rotundatum*)
439	Sep 3	Doral NW 53rd St., Miami	25 49'22.08" N 80 22'30" W	1 nymph *A. rotundatum*
451	Sep 3	Florida International Univ., Pond 2	25 46'7.68" N 80 21' 46.08" W	4 larvae *Amblyomma* spp. (most likely *A. rotundatum*)
461	Sep 3	Florida International Univ., Pond 1	25 46'7.68" N 80 21' 46.08" W	1 larva *Amblyomma* spp. (most likely *A. rotundatum*)

Material Examined. 12 nymphs, 14 larvae, Dade County, Florida: Doral NW 53rd St., Miami, 09-V-2014, J. Dagg, ex. 6 cane toads, *Rhinella marina*; 5 larvae, Dade County, Florida: Florida International University, Miami, 3-IX-2014, J. Dagg, ex. 2 cane toads, *Rhinella marina*.

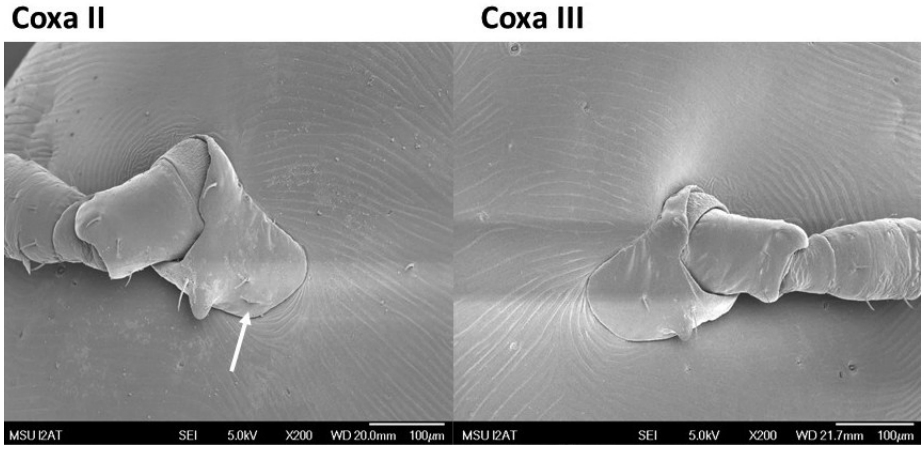

FIGURE 1. Coxae II and III of *Amblyomma rotundatum* removed from cane toads, Miami, FL. Arrow points to tiny internal spur on coxa II.

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