

## ***Ixodes downsi* Kohls, 1957 in Venezuela**

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### **Abstract**

Three females and three nymphs of *Ixodes downsi* Kohls, 1957 were collected in guano deposited in Guácharos Cave, State of Monagas, Venezuela. The cave is inhabited by bats and oilbirds (*Steatornis caripensis* Humboldt) and is frequented by carnivores. Previous records of this tick are from Trinidad & Tobago and Peru, and it is unclear whether the principal hosts are Chiroptera or Aves. The present record of *I. downsi* in Venezuela does not provide new host data but expands this tick's known range, which is probably greater than collections indicate. Phylogenetic analyses based on 16S mitochondrial rDNA sequences of *Ixodes* species from the Neotropical Zoogeographic Region, plus two Australian *Ixodes* and three argasids, were conducted to clarify the systematic position of *I. downsi*. The analyses showed an unexpected relationship with *I. (Multidentatus) auritulus* Neumann, 1904. However, as with other Neotropical *Ixodes*, the subgeneric status of *I. downsi* remains undetermined.

**Key words:** Ixodidae, *Ixodes downsi*, Chiroptera, Aves, cave, 16S sequences, Venezuela

### **Introduction**

*Ixodes downsi* Kohls, 1957 is a Neotropical species described from a larva collected on *Anoura geoffroyi* Gray, 1838 (Chiroptera: Phyllostomidae) and nymphs, males and females collected in the same cave as the infested bat in Cerro Aripo (10° 43' N 61° 15' W), Trinidad, Trinidad & Tobago. The only additional record of *I. downsi* is that of Wilson & Baker (1988), who found a female on a “guácharo,” *Steatornis caripensis* Humboldt, 1814 (Aves: Caprimulgiformes: Steatornithidae), at a cave in Tingo María (09° 17' S 75° 59' W), Huanuco Region, Peru. Herein we report the first record of *I. downsi* from Venezuela and discuss this tick's relationship with other Neotropical species by comparing sequences of the mitochondrial 16S rDNA gene.

### **Materials and methods**

Six ticks belonging to the genus *Ixodes* were collected on May 15, 2007 from guano deposited in Cueva del Guácharo (10° 11' N 63° 42' W), State of Monagas, Venezuela. The cave is located in Parque Nacional El Guácharo, which comprises more than 62,000 ha in the states of Monagas and Sucre. Ticks were identified using keys and descriptions for Neotropical *Ixodes*. The cave is inhabited by several species of Chiroptera and also contains a population of *S. caripensis*. Carnivora, chiefly Mustelidae, frequent the cave to prey upon bats and birds.

Two ticks (a female and a nymph) were preserved in ethanol and stored at  $-20^{\circ}\text{C}$  until they could be used for DNA extraction and polymerase chain reaction (PCR) amplification, as described by Mangold *et al.* (1998), to obtain sequences of 16S rDNA. This sequence was added to the sequences used in Guglielmone *et al.* (2006), namely, sequences of Neotropical *Ixodes* deposited in GenBank plus sequences of the Australian *I. holocyclus* Neumann, 1899 and *I. tasmani* Neumann, 1899. Sequences of *Argas persicus* (Oken, 1818), *Ornithodoros moubata* (Murray, 1877) and *Otobius megnini* (Dugès, 1883) were used as outgroups. See Guglielmone *et al.* (2006) for GenBank accession numbers of sequences for the different species.

A neighbor-joining condensed tree was generated using the Kimura two-parameter distance method, and gaps were excluded in the pairwise comparison to determine the relationship of the specimens from Guácharo Cave with other Neotropical *Ixodes*. Maximum parsimony analysis was also performed using Mega 4.0 and the heuristic search procedure (Tamura *et al.*, 2007). Tick specimens used for DNA extraction have been deposited in the tick collection (DNA section) of the Instituto Nacional de Tecnología Agropecuaria, Estación Experimental Agropecuaria Rafaela, Rafaela, Santa Fe, Argentina, and the remaining ticks have been deposited in the tick collection of the Departamento de Ciencias Pecuarias, Laboratorio de Zoología, Facultad de Medicina Veterinaria in Chillán, Chile.

## Results

The entire tick collection consisted of three females and three nymphs. One female was partially engorged, and the remainder were unfed. The females were characterized by hirsute body (dorsally and ventrally), broadly triangular prosoma with sinuous posterior dorsal border and short, blunt cornua and auriculae; and large porose areas, wider than long and separated by much less than the diameter of one area. Hypostome blunt with 3/3 denticles apically (first row), then 2/2 to end of toothed portion. Scutum oval with few, scattered small punctations; setae anteriorly and in antero-lateral fields. Coxae I–IV with short, triangular, blunt external spurs (largest on coxa I). Trochanters without spurs. Spiracular plate sub-oval. Nymphs similar to females but with fewer setae on body and scutum; spiracular plates sub-oval with goblets fewer and larger than in females; and posterior dorsal border of prosoma straight. These features and published descriptions of Neotropical *Ixodes* led us to conclude that all our specimens are *I. downsi* Kohls, 1957.

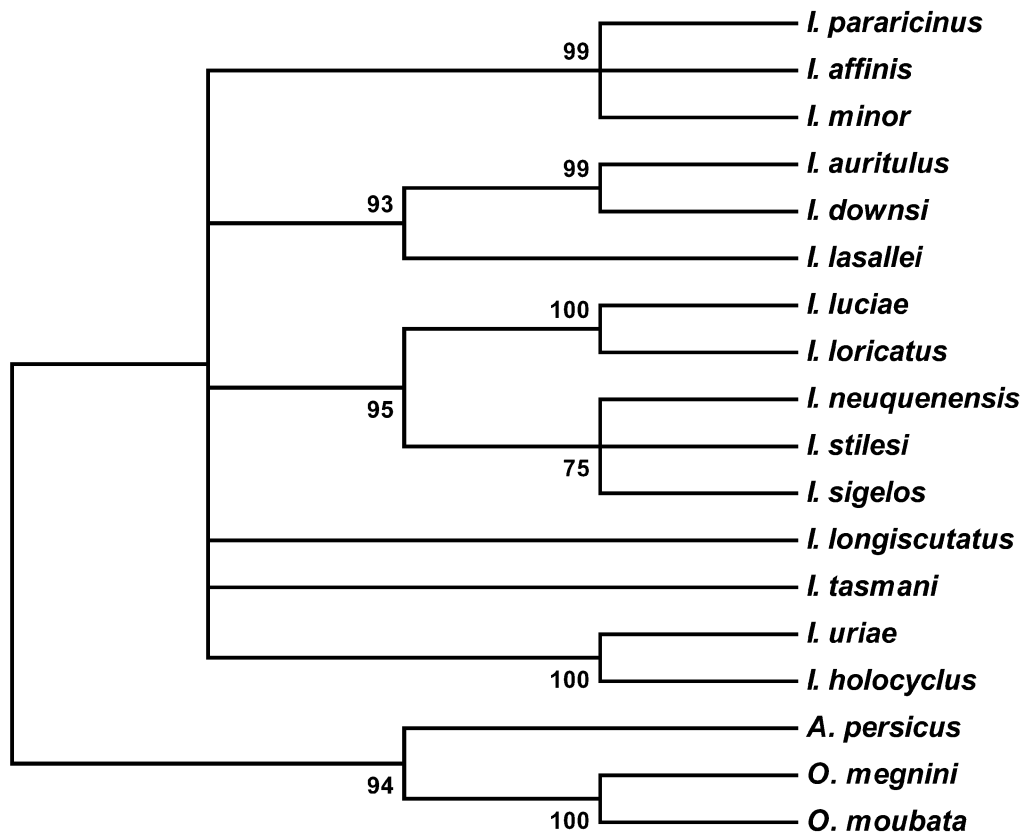
The GenBank accession number for the female *I. downsi* 16S sequence is EU195917, and that for the nymph is EU195918. The neighbor-joining tree (Figure 1) indicates that *I. downsi* forms a clade with *I. auritulus* Neumann, 1904 and *I. lasallei* Méndez Arocha and Ortiz, 1958. However, maximum parsimony confirmed only a clade formed by *I. auritulus* and *I. downsi* (95% support, data not shown).

## Discussion

*Ixodes downsi* has been collected from bats and oilbirds inhabiting caves (Kohls, 1957; Wilson & Baker, 1988). The current record of this tick species in Venezuela does not add to our knowledge of host specificity but serves to expand this tick's known range, which is probably greater than the few available collections have indicated. The finding of a partially engorged female in guano was unexpected. It was probably a consequence of detaching from a dead host.

The subgeneric status of *Ixodes downsi* was considered uncertain by Clifford *et al.* (1973). The 16S rDNA sequence data presented here do not alter this diagnosis, because neighbor-joining tree

topology shows that *I. downsi* forms an unexpected clade with *I. auritulus* and *I. lasallei* [other clades are similar to those presented in Guglielmone *et al.*, (2006)]. Nevertheless, the maximum parsimony method reduced the clade to *I. downsi* and *I. auritulus*. The relationship between these two species is not obvious. According to Clifford *et al.* (1973), *I. auritulus* (a bird tick) is a member of the subgenus *Multidentatus*, whose chief character is a highly denticulate hypostome, 5/5 or 6/6 in females and 3/3 or 4/4 in nymphs. Consequently, additional molecular and morphological studies on *Ixodes* from the Neotropical Zoogeographic Region are needed to better define natural groups within the genus *Ixodes*.



**FIGURE 1.** Neighbor-joining condensed tree using the Kimura two-parameter distance method. Numbers on branches represent bootstrap support (more than 50%) generated from 1,000 replications.

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