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ORIGINAL ARTICLE

Presence of *Cooperia curticei, C. punctata* and *Trichostrongylus colubriformis,* (Strongylida: Trichostrongylidae) in Tabasco, Mexico

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ABSTRACT: The aim of this study was to report and morphologically describe two small intestinal nematode genera present in cattle and sheep from Tabasco, Mexico. The specimens studied were obtained from animals for slaughter on a trail, cleared in Amann's lactophenol and their measurements were recorded using a calibrated ocular micrometer. Based on their morphologic characteristics, three species were identified: *Cooperia curticei*, *Cooperia punctata* and *Trichostrongylus colubriformis*. In *C. curticei,* the length of the tail-vulva was higher (1726.9± 194.4 µm) than in *C. punctata* (1422.1±151.7 µm); and the vulva shape of *C. curticei* was flat, while in *C. punctata,* it was botonous. In *Cooperia* males, the spicule lengths were similar in both species (139±13 µm). The differentiation was carried out morphologically; *C. curticei* and *T. colubriformis* were the species found in the small intestine of sheep, while *C. puntata* was found in that of cattle.

Key words: *Cooperia curticei*, *Cooperia punctata*, *Trichostrongylus colubriformis*, ruminants, parasites.

Presencia de *Cooperia curticei, Cooperia punctata* y *Trichostrongylus colubriformis,* (Strongylida: Trichostrongylidae) en Tabasco, México

RESUMEN: El objetivo del estudio fue notificar la presencia, y describir morfológicamente, dos géneros de nematodos encontrados en intestino delgado de bovinos y ovinos en Tabasco, México. Los nematodos adultos se obtuvieron de animales para abasto en un matadero; se aclararon con lactofenol de Amman y se registraron sus medidas mediante un micrómetro ocular calibrado. Según las características morfológicas, se identificaron tres especies: *Cooperia curticei*, *Cooperia punctata* y *Trichostrongylus colubriformis*. Se observó que la longitud de la punta de la cola a la vulva fue mayor en *C. curticei* (1726.9± 194.4 µm) que en *C. punctata* (1422.1±151.7 µm) y la forma de la vulva en *C. curticei* fue plana, mientras que en *C. punctata* fue de tipo botonosa. En los machos de *Cooperia* las medidas de las espículas fueron similares entre las dos especies (139±13 µm) y se diferenciaron sólo por su morfología. *C. curticei* y *T. colubriformis* fueron las especies encontradas en intestino delgado de ovinos y *C. punctata* en intestino delgado de bovinos.

Palabras clave: *Cooperia curticei*, *Cooperia punctata*, *Trichostrongylus colubriformis*, rumiantes, parásitos.

INTRODUCTION

Diagnostic tests play an important role in confirming the presence of gastrointestinal nematodes (GIN) in ruminants (1). Many epidemiological studies are based on nematode egg counts in faeces, morphologic identification to gender and sometimes to species in larvae from pasture and coproculture, while the confirmation of the species takes place in adult nematodes at the necropsy of animals (2). The small intestinal nematodes *Cooperia curticei* and *Trichostongylus colubriformis* has been identified in previous study, in Tabasco Mexico using the methologic characteristics (2, 3), while in Brazil, the same species

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are reported as strongly adapted to sheep (4) but morphometric characteristics are not reported.

In recent years, there have been some advances towards the development of molecular diagnostic tools for identifying GIN that commonly infect ruminants (1, 5, 6), but that is not available in all local institutions in which diagnosis to regional studies is required, especially when gastrointestinal nematode parasites remain as one of the major parasitic disease to ruminant production in the tropics (7). For this reason, traditional diagnostic techniques help identifying the nematode species, so the aim of this study was to report and describe morphologically two small intestinal nematode species present in cattle and sheep in Tabasco, Mexico.

MATERIALS AND METHODS

Adult specimens of intestinal nematodes of sheep were obtained from a slaughterhouse in Villahermosa, Tabasco, while *C. punctata* was obtained from Zebu cattle slaughtered in Teapa, Tabasco. In the most places of the sheep origin, climate (Af or Am) is hot and humid, with abundant rains in summer and average temperature between 23.8 and 25.8°C (8).

Nematode collection: The small intestine was tied off into 3 m sections and washed with saline solution to collect adult nematodes. Each volume of solution was forced through the intestine by applying pressure between thumb and finger. Samples were taken to the Laboratory of Regional Universitary Sursureste Unit (URUSSE), belonging to the Autonomous University of Chapingo (UACh), to be processed. The small intestine was washed with tap water in a 400 mash (0.038 mm, Mont-inox). Specimens were cleared studied in Amann's lactophenol (2) and subsequently measurements were recorded using a calibrated ocular micrometer. Body and esophagus lengths were measured, except in *C. curticei*. In the males, spicules length was recorded. In females, the anterior and posterior sphincter, vestibule and tip tail-vulva lengths were measured. The test was performed to determine the differences in morphology between the two species of *Cooperia* (9). Specific identification of males was carried out by a morphologic key (10). Photographs were obtained with a Canon Power Shot-A 400, 3.2 Mega pixels. Digitized images were selected and edited in the Fire Works software.

RESULTS AND DISCUSSION

Two *Cooperia* species *C. curticei* in sheep and *C. puntata* in cattle were identified. In females they had slow differences in the vulva shape as shown in Figure 1,

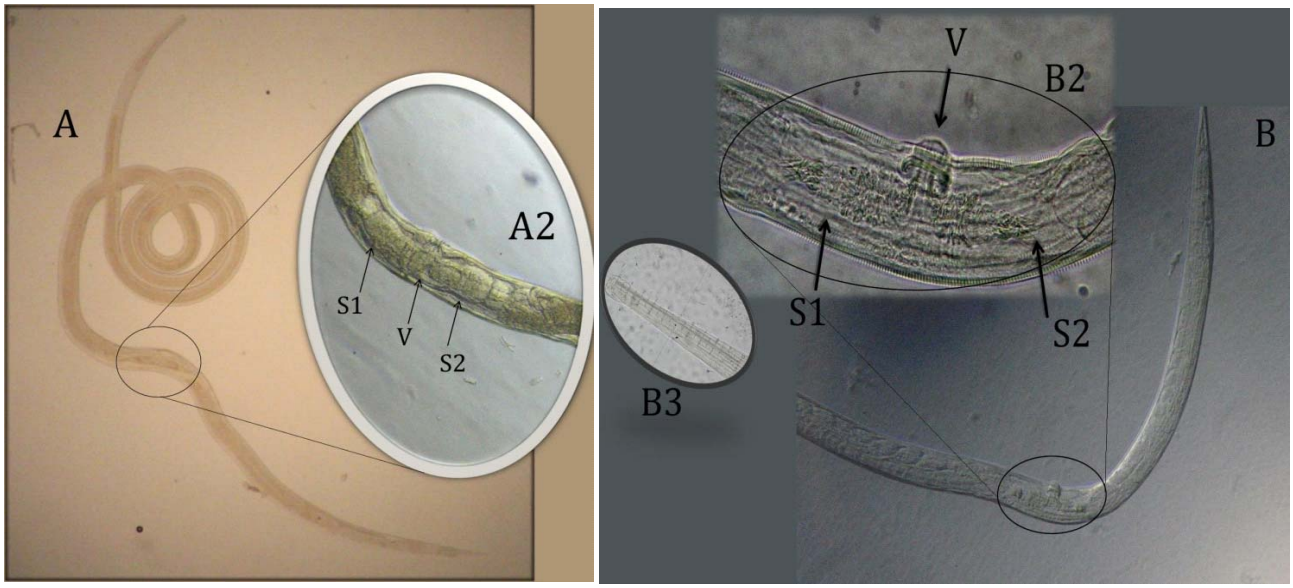

FIGURA 1. *Cooperia punctata* females (right, zebu cattle) and *Cooperia curticei* (left sheep). S1. Anterior sphincter, S2. Posterior sphincter, V. Vestivule-vulva. B3. Anterior end./ *Hembras de* ***Cooperia punctata*** *(derecha, ganado cebú) y* ***Cooperia curticei*** *(izquierda, ovino). S1. Esfinter anterior, S2. Esfinter posterior, V. Vestíbulo-vulva. B3. Parte anterior.*

C. curticei showed a flatted vulva, while in *C. punctate,* it was botonous. Also those species showed differences in sizes ($p<0.01$). Higher tail-vulva length was recorder in *C. curticei* (1726.9 µm) while *C. punctata* was slightly smaller (1422.1 µm).

The resume of the main morphological aspects measured in the three nematode species is show in Table 1. Females are not generally used to identify species, there is little information regarding morphological measures.

The morphometry of spicules in males of *C. curticei* and *C. punctata* was similar ($p>0.05$) (Table 2), and differentiation was carried out by morphology as suggested by Stringrellow (10). The results obtained from the morphometry of spicules were similar to other studies carried out in *C. curticei* (11, 12).

C. curticei spicules showed a ventral flange, without concavity, distally spicule sharply curved medially. *C. punctata* had a large concavity near the middle of spicule, border of concavity projects laterally at an angle from spicule shaft, ventral flange posterior to concavity not pronounced (10). An easy way to distinguish *C. curticei* is the rolled shape taken when fixed in formalin (Fig. 2).

In *T. colubriformis,* the spicules are slightly unequal in length, with a structure similar to a small boat with a thick outgrowth capping the root proximally (Figure 3). *T. colubriformis* corresponds to the description given by other authors (13, 14). The principal difference between *T. colubriformis* and *Cooperia* spp. was the head shape and the presence of the excretory pore in *T. colubriformis* (Figure 3).

TABLA 1. Morphometry of female nematodes of sheep and cattle small intestine./ *Morfometría de hembras de nematodos de intestino delgado de bovinos y ovinos.*

Character	Sheep				Cattle	
	Cooperia curticei		*Trichostrongylus colubriformis*		*Cooperia punctata*	
	Average	SD	Average	SD	Average	SD
Number measured	77		21		18	
Body length	*		5.4	0.6	5.5	0.7
Esophagus length (µm)	325.8	23.4	774.3	105.4	298.5	68.1
Anterior sphincter length (µm)	109.6	29.9	139.4	20.3	105.4	23.4
Vestibule length (µm)	87.3[a]	39.3	123.5	37.7	30.9[b]	7.1
Posterior sphincter length (µm)	106.8	25.5	151.3	34.4	108.7	21.5
Tail-vulva length (µm)	1726.9[a]	194.4	1249.5	119.4	1422.1[b]	151.7
Tail length (µm)	142.2	23.0	83.2	29.3	158.3	5.0
Number of eggs in uterus	29.2	9.1	18.5	6.9	23.1	2.1

*It was not possible to measure because it was enrolled. Different letters between the two *Cooperia* species indicate significant differences ($P<0.01$).

TABLA 2. Morphometry of male nematodes of sheep and cattle small intestine./ *Morfometría de nematodos machos de intestino delgado de bovinos y ovinos.*

Character	Sheep				Cattle	
	Cooperia curticei		*Trichostrongylus colubriformis*		*Cooperia punctata*	
	Average	SD	Average	SD	Average	SD
Number measured	91		36		26	
Body length	4.9	0.9	5.3	0.8	5.1	1.1
Right spicule	138.67	13.42	140.41	10.97	136.22	6.98
Left spicule	140.35	13.53	144.45	15.37	134.85	5.79

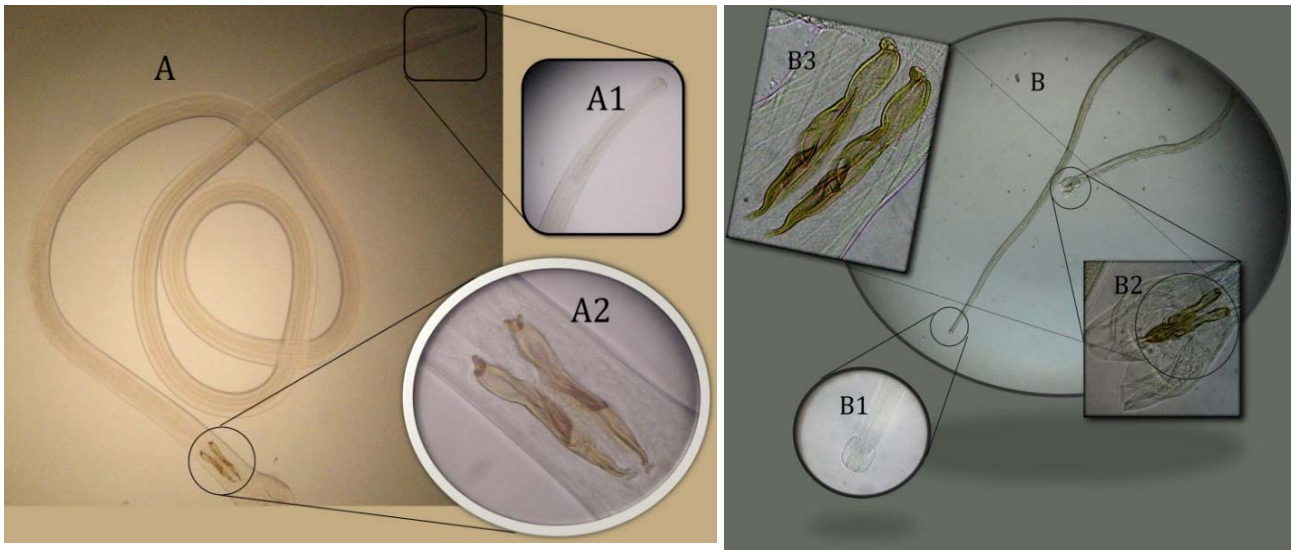

FIGURA 2. Males of *Cooperia curticei* (left, sheep) and *Cooperia punctata* (right, Zebu cattle). B2. Copulatrix bursa, A2 and B3. Spicules, A1 and B1. Anterior end./ *Machos de **Cooperia curticei** (izquierda, ovinos) y **Cooperia punctata** (derecha, ganado Cebú). B2. Bursa copulatriz, A2 y B3. Espículas, A1 y B1. Parte anterior.*

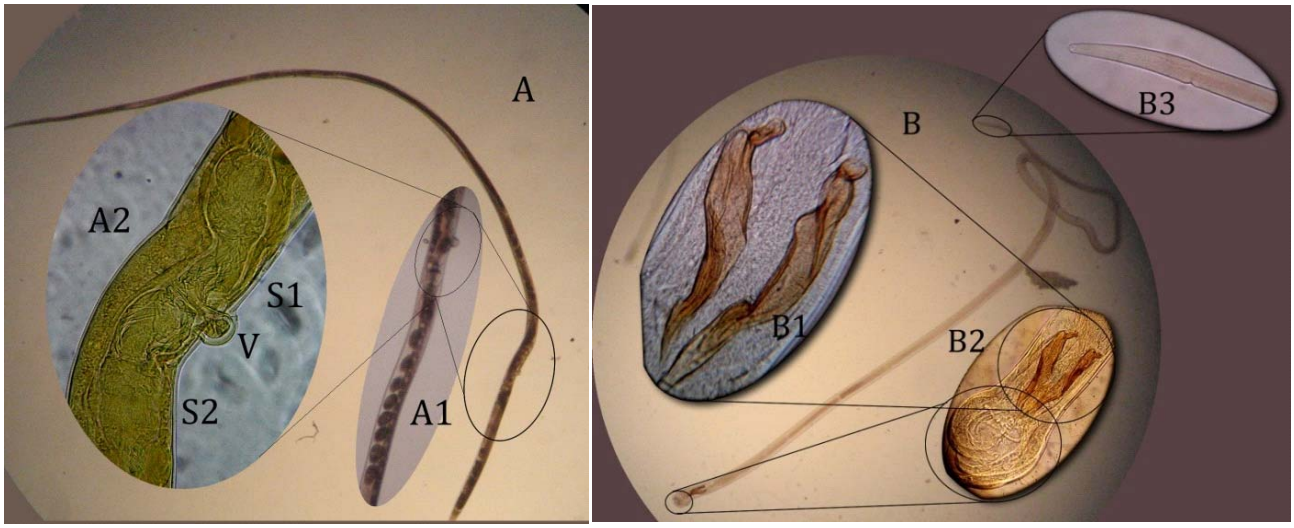

FIGURA 3. Nematodes of *Trichostrongylus colubriformis*. Female left (A). S1. Anterior sphincter, S2. Posterior sphincter, V. Vestivule-vulva. Male right (B). B1. Spicules, B2. Copulatrix bursa, B3. Anterior end and excretory pore./ *Nematodos de **Trichostrongylus colubriformis**. Izquierda hembra (A), S1. Esfí5nter anterior, S2. Esfínter posterior, V. Vestíbulo-vulva. Derecha macho (B), B1. Espículas, B2. Bursa Copulatriz, B3. Parte anterior con poro excretor.*

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