CYSTOOPSIS ATRACTOSTEI N. SP. (NEMATODA: CYSTOOPSIDAE) FROM THE SUBCUTANEOUS TISSUE OF THE TROPICAL GAR, *ATRACTOSTEUS TROPICUS* (PISCES), IN MEXICO

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ABSTRACT: A new species of parasitic nematode, *Cystoopsis atractostei* (Trichinelloidea: Cystoopsidae), is described based on female specimens recovered from the subcutaneous tissue of the tropical gar, *Atractosteus tropicus* Gill (Lepisosteiformes: Lepisosteidae), from 2 localities (Canal Nueva Esperanza and Canal Tabasquillo) of the Pantanos de Centla Biological Reserve, State of Tabasco, southeastern Mexico, collected in April 2001. The total prevalence was 13%, and the mean intensity of infection was 1 nematode per fish. The new species differs from females of the only other adequately described congeneric species, *C. acipenseris* Wagner, 1867, mainly in possessing a bulbous inflation at the anterior end of the muscular esophagus, the vulva situated well posterior to the nerve ring, smooth cuticle, and in the shape of the posterior vesicular portion of the body (markedly transversely oval) in the largest specimens. Both species also differ in their host types (Lepisosteiformes vs. Acipenseriformes) and in geographical distribution (tropical southern Mexico vs. temperate zones of the Holarctic).

The trichinelloid *Cystoopsis* Wagner, 1867, comprises nematodes with marked sexual dimorphism. Whereas the body of the male is cylindrical, that of the female is divided into an anterior cylindrical, filiform region and a posterior globular region filled with uterine coils. Until recently, the genus has been represented by a single species, *Cystoopsis acipenseris* Wagner, 1867, a widely distributed parasite of the skin of the Holarctic sturgeons (Acipenseridae) (Moravec, 2001a). However, Osorio-Sarabia et al. (1987) reported *Cystoopsis* sp. in the tropical gar *Atractosteus tropicus* Gill, in the State of Tabasco, southeastern Mexico.

In April 2001, investigations into the helminth parasites of tropical gars were carried out in Tabasco, oriented particularly to obtaining new data on this interesting nematode. Examinations of gars from 3 localities of the Pantanos de Centla Biological Reserve yielded a few female specimens of *Cystoopsis*, which made it possible to study their morphology in more detail and to describe a new species.

MATERIALS AND METHODS

In April 2001, 38 tropical gars, *A. tropicus* Gill (total body length, 33–63 cm), were examined for the presence of *Cystoopsis*. They were caught by professional fishermen in the following 3 localities in the Pantanos de Centla Biological Reserve: (1) Canal Nueva Esperanza (18°23'78"N, 92°34'78"W) (22 specimens); (2) Canal Tabasquillo (18°14'12"N, 92°49'25"W) (9 specimens); and (3) San Isidro Lake (18°21'55"N, 92°29'88"W) (7 specimens). The fish were examined so that first the external, visual examination of the body surface was done to find possible nodules caused by the parasite. Then, the broad bands of the skin, including bony scales, were removed from the whole body surface of fish with the aid of scissors and a scalpel, and the inner surface of them was observed in petri dishes with physiological saline under a dissecting microscope. The parasites were removed from the surrounding tissue with the help of fine mounting needles and were then fixed in hot 4% formaldehyde.

In addition to fishes from the Pantanos de Centla Biological Reserve, the body surfaces of 25 tropical gars in the fish market in Villahermosa were examined visually for the presence of *Cystoopsis* nodules, but none was found.

For examination, the nematodes were cleared in glycerine. Drawings were made with the aid of an Aristoplan microscope drawing attach-

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DESCRIPTION

Cystoopsis atractostei n. sp. (Fig. 1, 2)

Description of female (based on 5 specimens; measurements of holotype in parentheses): Body divided into 2 parts: anterior filiform part 3.5-4.8 mm (3.5 mm) long, 63-66 (66) wide; posterior vesicular part transversely oval to almost spherical, 1.99-2 mm (2 mm) long, 2.58-4.8 mm (4.8 mm) wide. Cuticle of anterior portion of body appearing smooth, without transverse rings of tubercles. Mouth aperture surrounded by ring mound 6 (6) long, 30 (30) wide; cephalic papillae indistinct. Anterior muscular part of esophagus well developed, 465-492 (492) long; its anterior end distinctly inflated, bulbous, 39-42 (42) long, and 48 (48) wide; width of muscular esophagus just posterior to bulb 27 (27), its maximum width at posterior region 33 (33). Posterior glandular esophagus (stichosome) formed by 2 rows of small cells (stichocytes), extending posteriorly to end of filiform part of body. Intestine ending blindly, forming sac 530-884 (not measured in holotype) long and 136-163 wide, situated in posterior vesicular part of body. Nerve ring just posterior to anterior esophageal bulb, 60-66 (60) from anterior extremity. Excretory pore 171-186 (186) from anterior extremity. Vulva with somewhat elevated lips, 93-105 (105) from anterior end of body. Muscular vagina passing through entire filiform region of body. Uterine convolutions along with ovary filling body cavity in posterior part of body. Uterus containing large number of eggs at different stages of development; mature eggs containing formed larva. Young eggs, as compared with more developed eggs, with thinner walls, polar plugs distinctly protruding. Two types of mature eggs differing markedly in size: (1) mature eggs of holotype, 2 paratype specimens large, 60-63 long, 21-23 wide; egg wall 2 thick; polar plugs 4 long, 6 wide; and (2) mature eggs of 2 other paratypes small, 36-42 long, 15-18 wide; egg wall 2-3 thick; polar plugs 3 long, 4-6 wide.

Male: Unknown.

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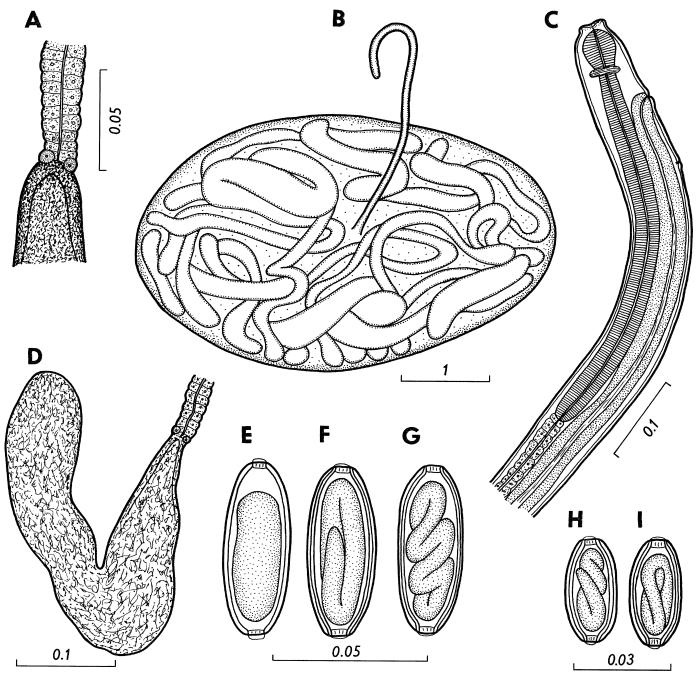


FIGURE 1. *Cystoopsis atractostei* n. sp., gravid female. A. Region of esophago-intestinal junction. B. General view. C. Anterior end of body. D. Intestine. E–G. Large eggs at different stages of development. H, I. Small eggs. Scale bars in millimeters.

Taxonomic summary

Type host: Tropical gar, *Atractosteus tropicus* Gill, 1863 (Lepisosteidae, Lepisosteiformes) (total body length, 33–51 cm).

Site of infection: Under skin (scales).

Type locality: Canal Nueva Esperanza (18°23'78"N, 92°34'78"W), Pantanos de Centla, State of Tabasco, Mexico (holotype collected on 26 April 2001).

Other locality: Tabasquillo (18°14′12″N, 92°49′25″W), Pantanos de Centla, State of Tabasco, Mexico (27 April 2001).

Prevalence and intensity: Canal Nueva Esperanza-18% (4

fish infected per 22 fish examined), 1 nematode per fish; Tabasquillo—11% (1 fish infected per 9 fish examined), 1 nematode per fish. No *Cystoopsis* was recorded in 7 *A. tropicus* examined from San Isidro Lake. Total prevalence in Pantanos de Centla was 13%; mean intensity, 1.

Deposition of types: Holotype and paratype in the Institute of Biology, National Autonomous University of Mexico (UNAM), Mexico City (Cat. Nos. 4439 and 4440). Paratypes in the helminthological collection of the Institute of Parasitology, Academy of Sciences of the Czech Republic, in České Budějovice (Cat. No. N-792).

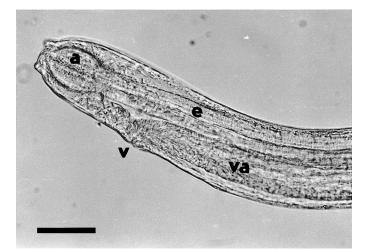


FIGURE 2. *Cystoopsis atractostei* n. sp., anterior end of female (holotype). a, anterior inflation of esophagus; e, esophagus; v, vulva; va, vagina. Scale bar = $40 \ \mu$ m.

Etymology: The specific name of this nematode relates to the generic name of the fish host.

DISCUSSION

To date, *Cystoopsis* Wagner, 1867, the only genus in the trichinelloid Cystoopsidae Skryabin, 1923, has contained only 1 well-described species, *C. acipenseris* Wagner, 1867, a parasite in the skin of sturgeons (Acipenseridae) in Europe, Palaearctic Asia, and North America (Moravec, 2001a). The second nominal species, *C. scomber* Zlatev, 1936, poorly described from the inner surface of gill covers of the marine fish *Scomber scombrus* Linnaeus (Scombridae, Perciformes) in Bulgaria (Zlatev, 1936), was designated a species inquirenda by Moravec (2001a); he mentions that, in fact, this parasite might be a didymozoid trematode.

Even though no male of C. atractostei n. sp. was obtained, its gravid females can be differentiated morphologically from those of C. acipenseris in having a bulbously inflated anterior end of the muscular esophagus, which is distinctly broader than the posterior part of the muscular esophagus (vs. anterior end of muscular esophagus noninflated, somewhat narrower than the posterior part of esophagus), in the vulva situated distinctly posterior to the nerve ring (vs. anterior to nerve ring-see illustration by Lomakin in Bauer, 1987), and the vesicular portion of body of the large female being markedly transverse oval (vs. spherical). The cuticle of the filiform part of the body of C. atractostei appears to be smooth, whereas that in C. acipenseris is covered with minute tubercles arranged in regular transverse rings. The intestinal sac of C. atractostei is much smaller than that of C. acipenseris (less than 1-mm long vs. 2-2.5 mm in diameter).

The size of mature eggs of *C. acipenseris* is reported to be $63-68 \times 21-25 \ \mu\text{m}$ (Moravec, 2001a), but there were 2 sizes of eggs of *C. atractostei*: large eggs measuring $60-63 \times 21-23 \ \mu\text{m}$ and small eggs measuring $36-42 \times 15-18 \ \mu\text{m}$ with usually somewhat thicker shells. The larger eggs seem to be associated with broader uterine coils. The larger eggs were found, in our opinion, in younger nematodes and, therefore, it

is possible that the size of produced eggs gradually decreases during the life of the nematode. Another explanation may be that the nematodes can contemporarily produce both types (production of 2 types of eggs is known in some other trichinelloids, e.g., *Paracapillaria philippinensis* (Chitwood, Velasquez, and Salazar, 1968) or *Trichosomoides crassicauda* (Bellingham, 1840) [see also Chitwood et al., 1968; Moravec, 2000, 2001b]), but this is not obvious from the present material. Nevertheless, despite the different sizes of eggs, all specimens of the present material are considered to belong to the same species.

In addition to morphological differences, *C. atractostei* and *C. acipenseris* differ in the host types (Lepisosteiformes vs. Acipenseriformes) and the geographical distribution (tropical region of southern Mexico vs. temperate zones of the Holarctic).

Cystoopsis atractostei was already recorded as Cystoopsis sp. from A. tropicus in Tabasco (Lake El Chiribital), southern Mexico, by Osorio-Sarabia et al. (1987), who gave a brief description and drawings of females. In contrast to the present findings, they illustrated the muscular esophagus without an anterior bulbous inflation and a distinctly smaller posterior vesicular part of the body. The reported size of eggs (96 × 80 µm) is evidently erroneous because the scale accompanying the drawing of the mature egg (their figure 10b) shows that the length of the egg slightly exceeds 30 µm.

Their specimens were deposited in the helminthological collection of the Instituto de Biología, UNAM, in Mexico City, but a recent reexamination of this material (Cat. No. 2203) showed that the vial contained only the uterine fragments with eggs. All the eggs examined were small, larvated, thin- or thickwalled, measuring $36-42 \times 18 \mu m$; the thickness of the wall was mostly less than 2 μm , but in some eggs, the thickness was up to 3 μm ; the polar plugs were 3- μm long and 4- to 6- μm wide, with the protruding part being $1-2 \mu m$; the width of the body of the larva was 4 μm . In spite of some morphological differences, which may be assigned to inaccurate observations, and considering that they were found in the same fish species in the same region, it is clear that the specimens described as *Cystoopsis* sp. by Osorio-Sarabia (1987) were conspecific with *C. atractostei*.

Osorio-Sarabia et al. (1987) report that these parasites were found in fibrous cysts in the host's epidermis and that the nodules caused by them were very marked on the host's body surface; this is confirmed by a fixed piece of the host's epidermis in the vial with *Cystoopsis* sp. deposited in UNAM, which forms an elevated protuberance at the site covering the parasitic cyst. Similar conspicuous nodules are known to occur on the skin of acipenserids infected with *C. acipenseris*. Therefore, it is interesting that no such nodule was observed on the skin of gars infected with *C. atractostei* from Pantanos de Centla, where the parasites were always located beneath the bony scales of the host. The external examination of 25 tropical gars in the fish market in Villahermosa, Tabasco, did not reveal any nodules on the skin of these fish.

According to Janicki and Rašín (1928, 1929, 1930), *C. acipenseris* occurs under the skin of sturgeons inside the cysts (mainly between bony scales), where 1 cyst always contains 1 male and 1 female parasite; after the cyst ruptures, the parasite's uterine coils are liberated and they burst to release the nematode's eggs into the water. In contrast, we observed *C. atractostei* females embedded rather deeply under the skin between

bony scales inside a very fine capsule, not accompanied by a male. Although the rupture of the female body after completion of the biological cycle cannot be excluded, the shape of the female body and the situation of the vulva near the cephalic end suggest that, at the time when eggs are mature, the female penetrates with its anterior filiform part of body through the host's skin to lay eggs into the external environment. The intermediate hosts of *C. acipenseris* are freshwater amphipods (Janicki and Rašín, 1930), and probably the same can be expected for *C. atractostei*.

Hosts of both *C. acipenseris* and *C. atractostei* belong to ancient groups of fishes, Acipenseriformes and Lepisosteiformes, and it may be assumed that these parasite species are also very old. However, in addition to the above species, Hoffman (1999) has recently reported on the finding of *Cystoopsis* sp. from centrarchids *Lepomis auritus* (Linnaeus) and *L. macrochirus* Rafinesque in Alabama and Georgia, by Cochran (1979), and the finding of *Cystoopsis* sp. from the African cichlid aquarium fish *Trophaeus moorei* (? = *Pseudochromis moorei* Fowler) (female not spheroid!) (G. Hoffman, unpubl. data). However, these records need verification as to whether these nematodes actually belonged to *Cystoopsis*.

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LITERATURE CITED

- BAUER, O. N. (ED.). 1987. Key to parasites of freshwater fishes of the fauna of the USSR. Part 3. Nauka, Leningrad, Russia, 583 p. [In Russian.]
- CHITWOOD, M. B., C. VELASQUEZ, AND N. G. SALAZAR. 1968. Capillaria philippinensis sp. n. (Nematoda: Trichinellida), from the intestine of man in the Philippines. Journal of Parasitology 54: 368–371.
- COCHRAN, M. L. 1979. Nematode parasites in freshwater fishes of Alabama and Georgia. Georgia Academy of Sciences 37: 199–204. [Ex Hoffman, 1999.]
- HOFFMAN, G. L. 1999. Parasites of North American freshwater fishes, 2nd ed. Cornell University Press, Ithaca, New York, 539 p.
- JANICKI, C., AND K. RAŠÍN. 1928. Die experimentelle Bestimmung des Zwischenwirtes von Cystoopsis acipenseri des Wolga-Sterlets. Die Naturwissenschaften 16: 821.
- —, AND —, 1929. Über die Entwicklung von Cystoopsis acipenseri N. Wagner 1867 im Zwischenwirt. Raboty Volzhskoy Biologicheskoy Stantsii 10: 187–209.
- —, AND —, 1930. Bemerkungen über Cystoopsis acipenseri des Wolga-Sterlets, sowie über die Entwicklung dieses Nematoden im Zwischenwirt. Zeitschrift für wissenschaftliche Zoologie 136: 1–37.
- MORAVEC, F. 2000. Review of capillariid and trichosomoidid nematodes from mammals in the Czech Republic and the Slovak Republic. Acta Societatis Zoologicae Bohemicae **64:** 271–304.
 - 2001a. Trichinelloid nematodes parasitic in cold-blooded vertebrates. Academia, Praha, Czech Republic, 429 p.
- 2001b. Redescription and systematic status of *Capillaria phi-lippinensis*, an intestinal parasite of human beings. Journal of Parasitology 87: 161–164.
- OSORIO-SARABIA, D., R. PINEDA-LÓPEZ, AND G. SALGADO-MALDONADO. 1987. Fauna helmintologica de peces dulceacuícolas de Tabasco. Estudio preliminar. Universidad y Ciencia 4: 5–31.
- ZLATEV, I. 1936. About some diseases of our fishes. Trudove na Chernomorskoy Biologitskoy Stantsii Varna 5: 67–79. [In Bulgarian, German summary.]