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SPIROXYS CONTORTUS PARASITISM ASSOCIATED WITH DOG BITE TRAUMA IN A CAPTIVE RED-EARED SLIDER (TRACHEMYS SCRIPTA ELEGANS) - A CASE REPORT

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ABSTRACT

A captive reared turtle (1-year-old male of Trachemys scripta elegans) was submitted for investigations (clinical, radiography, necropsy, parasitological and histological exams). The owner reported that the reptile was bitten by a 3-year-old male of Yellow Labrador retriever. Routine histological stain (hematoxylin-eosine) was used for parasites, stomach, intestine, liver and lungs. Parasitological investigations were focused on identification of the adult parasites. Preliminary clinical examination described fractures of the shell and concurrent hemorrhages. Radiological investigation revealed multiple fresh fractures of the shell and left clavicle, and viciously healed fracture of the left tibia and fibula. The most relevant lesions observed in gross investigations were the multiple fractures and characteristic dog bite perforations of the carapace and plastron. Hemorrhagic effusions in the coelomic cavity were remarked after the plastron removal produced by the liver rupture and lung perforation. The stomach presented gastric ulcerations and clustered coiled parasites from the Nematode class (Spiroxys contortus), heavily threaded into the gastric wall. Histological findings in liver revealed normal hepatic architecture. The hepatocytes presented vacuolated cytoplasm without modification of the central position of nuclei. The liver and lung presented zones of rupture filled with a clot bounded by a discrete fibrin layer and a small number of heterophils. Gastric wall presented healed ulcers and stage 3 larvae of Spiroxys contortus surrounded by giant multinucleated and mononuclear cells (parasitary granulomas).

Key words: dog bite, red-eared slider, Spiroxys contortus.

INTRODUCTION

The red-eared slider (*Trachemys scripta elegans)* is a semiaquatic turtle from the family *Emydidae,* being native to the Southern United States. It is the most popular pet turtle in this country and also in the rest of the world. The popularity of this species was created by the intense trade and also by the invasiveness which followed the pet releases (Shen et al., 2011). Captive omnivore turtles are fed with a variety of animals (earthworms, beetles, snails, shrimps, minnows) and vegetables (carrots, grapes, apples) (Bud, 2000). Considering the pathology, it is known that in turtles, trauma was the most frequent cause of morbidity and mortality, followed by death caused by unknown or undetermined agents, aural abscess, infectious diseases, and nutritional disorders (Brown and Sleeman, 2002). Dog bites are one of the most reported trauma in turtles, being frequently complicated with crush injury of the shell as a result of the high masticatory forces that can be delivered by certain breeds (Rosenthal, 1994; Presutti, 2001; Ciobotaru, 2013).

Regarding the helminthic fauna of turtles, there are only a few studies, especially in Romania, where the parasitism with the *Spiroxys contortus (*class Nematoda, family Gnathostomatidae) was found in European pond turtle *(Emys orbicularis)* (Mihalca et al., 2007; Miclaus et al., 2008).

The red-eared slider is known to carry nematodes (Hidalgo-Vila et al., 2009). Common helminthes which infect usually *Trachemys scripta elegans* are: *Serpinema microcephalus*, *Falcaustra donanaensis* and *Psysaloptera* spp. (Hidalgo-Vila et al., 2009), *Camallanus microcephalus, Oswaldocruzia leidy, Spiroxys constrictus, Spiroxys amydae, Spiroxys contortus, Cheloniadranculus* spp.

Aplectanus spp. (Ancholonu and Amy, 1971; Muzzall, 2005).

The last results show that 18 recognized species of *Spiroxys* have been reported from all zoogeographical regions of the world (Liang et al., 2014). *Spiroxys contortus* was found in Africa, Europe and North America in the stomach of turtles, following the ingestion of intermediate hosts such as copepod or other transport hosts (fish, amphibian). Adult nematodes of the genus *Spiroxys* are also parasites of the stomach and duodenum in other nonmarine chelonians, frogs, salamanders and colubrid snakes (Berry, 1985). The larvae penetrate mucosa, inducing granuloma formation in stomach and duodenal wall (Baker, 2007).

Experimentally, the indirect life cycle of *Spiroxys contortus* completes through the following stages: the incubation and hatching of the eggs into sheathed free-living larvae; development in artificially infected first intermediate hosts (species of copepods); the infection of second intermediate hosts (tadpoles); introduction of the larvae from the both intermediate hosts into the final hosts (turtles) and the growth of the larvae into adults (Hedrick, 1935).

The most infected reptiles with *S. contortus* are asymptomatic, only those with heavy burdens may become anorexic, underweighted, and develop melena and diarrhea (Baker, 2007).

The objective of this research was to describe grossly and microscopically, the lesions provoked by trauma (dog bite) and the gastric lesions associated with the incidental finding of *Spiroxys contortus,* this being the first case of infection in a captive red eared turtle.

MATERIALS AND METHODS

A captive turtle (1-year-old male of *Trachemys scripta elegans)* weighting around 1 kg was submitted for clinical investigations. The owner described that the reptile was bitten by a 3-year-old male of Yellow Labrador retriever. The death of the turtle occurred the next day after the bite, without any medication.

The turtle was submitted for ante mortem and postmortem investigations: clinical examination, radiography (55kVp, 5.0 mA), necropsy, parasitological and histological exams. Routine histological stain (hematoxylin-eosine) was used for adult nematodes, stomach, intestine, liver and lungs. Parasitological investigations were focused on identification of the adult parasites. Helminthes were counted, isolated and preserved in 70% ethanol and temporarily fixed with Amann lactophenol for identification.

RESULTS AND DISCUSSIONS

Preliminary examination of the turtle revealed fracture of the plastron and carapace. Multiple fresh fractures of the shell and left clavicle and viciously healed fractures of the left tibia and fibula were observed after radiography (Figure 1).

Figure 1. Red-eared slider, multiple fresh fractures of the shell and left clavicle (solid arrow), and viciously healed fracture of the left tibia and fibula (arrow)

The most relevant lesions observed during necropsy were the multiple fractures and characteristic dog bite perforations of the carapace and plastron (Figure 2, 3).

The distance between the perforations was 55 mm on carapace, 46 mm on plastron with a depth of approximately 1 cm.

During the necropsy, after the plastron removal there were observed hemorrhagic effusions in the coelomic cavity produced by the liver rupture and lung perforation (Figure 4).

Figure 2. Multiple fractures and characteristic dog bite perforations of the carapace

Figure 3. Multiple fractures and characteristic dog bite perforations of the plastron

The stomach presented gastric ulcerations and clustered coiled parasites of 2-3 cm from the Nematode class *(Spiroxys contortus),* heavily threaded into the gastric wall (Figure 5).

Figure 4. Liver rupture, deep tears with adherent fresh blood clots (arrow).

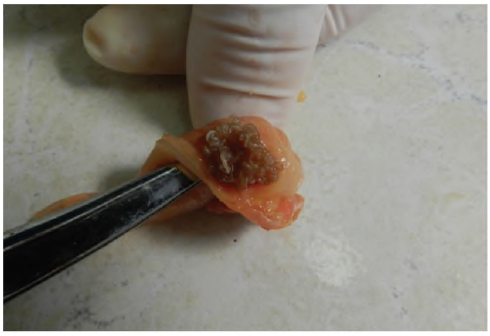

Figure 5. Stomach with clustered coiled adult parasites from the Nematode class *(Spiroxys contortus)*

The ulcers were pink, shallow, round shaped, with 3-5 mm in diameter.

Histological findings in liver revealed normal hepatic architecture. The hepatocytes presented vacuolated cytoplasm without modification of the central position of nuclei. The liver presented zones of rupture filled with a clot bounded by a discrete fibrin layer and a small number of heterophils (Figure 6). The same aspects were observed in the lungs.

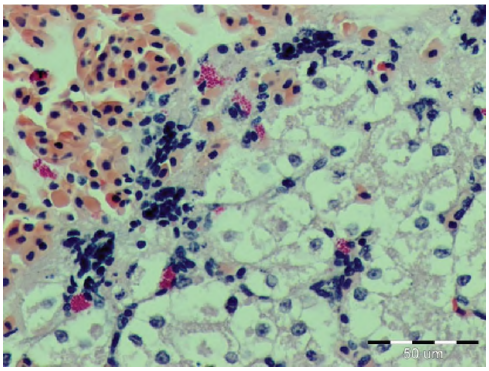

Figure 6. Liver with ruptures filled with erythrocytes and a small number of heterophils, hematoxylin - eosine (HE), x400

Ongoing healing ulcers and stage 3 larvae of *Spiroxys contortus* surrounded by giant multinucleated cells and mononuclear infiltrations numerically dominated by lymphocytes were identified in the stomach (parasitary granulomas - Figure 7). Larvae were also identified into the gastric lumen (Figure 8).

A number of 21 nematodes were counted after stomach opening: 16 of them being females and 5 males. The adults of *S. contortus*

measured 15 mm to 30 mm long. The mouth has two large, distinctly trilobed lips (Figure 9). The esophagus has a short anterior muscular portion and a longer posterior glandular portion. The male has well-developed caudal alae, delicate and subequal spicules, without gubernaculum. The vulva of the female is near the middle of the body, the ovary and uterus being well developed (Figure 10).

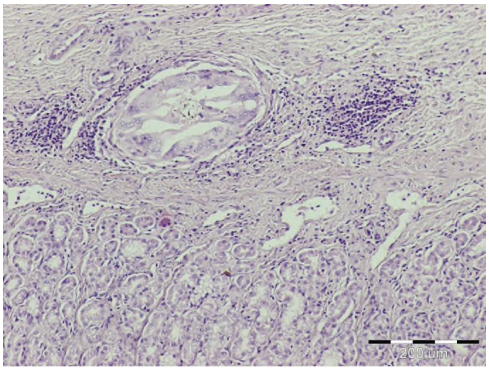

Figure 7. Gastric wall, small fragments of nematode surrounded by giant multinucleated cells and mononuclear infiltrate HE, x100

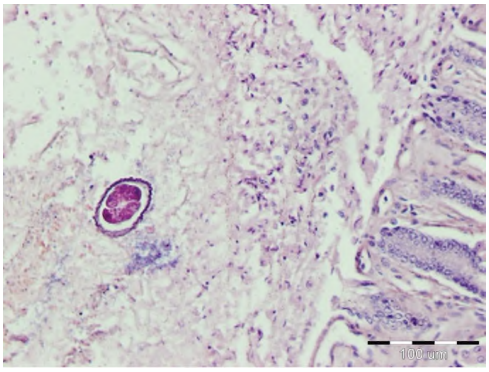

Figure 8. Gastric lumen with stage 3 larva of nematode, HE, x200

Data on the biology of turtle spirurines are poorly studied. The most encountered and described genera were *Spiroxys* and *Camallanus,* in which the life cycle involve aquatic arthropods (crustacean or insects) as intermediate hosts (Moravec and Vargas-Vasquez, 1998; Mascarenhas et al., 2013).

The life cycle of *Spiroxys contortus* starts with the eggs deposited by the females in the one- to two-

Figure 9. *Spiroxys contortus,* **anterior end with trilobed lips, x100**

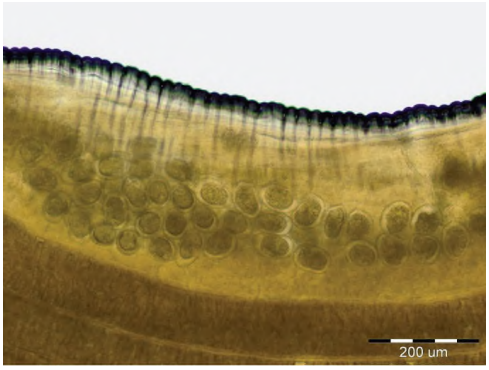

Figure 10. *Spiroxys contortus*, **female, lateral view of the intestine and uterus (eggs), x100**

cell stage. When passed in faeces of the turtle, they are in the 4- to 16-cell stage.

The eggs are fully developed in 84 hours in water at 23-25°C. The second stage larvae hatch within the cuticle of the first stage larvae, measuring 175-294 pm in length (Anderson, 2000). Larvae are readily ingested by *Cyclops albidus, C. brevispinosus, C. serrulatus* and *Mesocyclops leuckarti,* developing to the infective third stage in the haemocoel. Moulted larvae continue to grow slightly to the third stage in the copepod (Anderson, 2000). Considering the same studies, third stage larvae of *S. contortus* were found on the mesentery of various paratenic hosts namely mud minnows (*Umbra lima*), bullheads (*Ameiurus nebulosus*), tadpoles and frogs (*Rana clamitans*), larval and adult newts *(Triturus viridescens)* and dragonfly nymphs. These species can be considered as second natural intermediate host, enabling little growth of the larvae. Finally, the adult worms attach to the gastric wall of

various species of turtles, together with merely embedded or encysted larvae (Hedrick, 1935).
Wild turtles express the infection with *S. contortus* larvae during spring, adult worms being sporadically encountered before June. This nematode has an annual life cycle, starting as adult in the summer and continuing as larvae in winter (Anderson, 2000; Miclaus et al., 2008).
The owner declared that the turtle was bought from a pet shop and fed almost exclusively with fresh shrimps. Considering the life cycle particularly developed in this case, three possible scenarios can be discussed. The first include the shrimps as paratenic host for the larvae of *S. contortus*, the diet based on infected crustaceans being the way of infection. The second scenario refer to recent studies on the quality of drinking water (in treatment works, internal taps and hydrants), which is usually used for aquariums and aquaterariums. Thus, a diverse fauna was identified, both in European countries and United States (van Lieverloo et al., 2004; van Lieverloo et al., 2012). Copepodes, particularly *Cyclops albidus* and *Mesocyclops leuckarti* are frequently encountered in treated water (Evins, 2004). Thus, the presence of the first intermediate host in the water may create conditions for contamination of the turtle. This study does not include the results concerning the quality of the drinking water in Bucharest, but the possibility of contamination *via* drinking water cannot be excluded. The third scenario suspects that the contamination might be induced meanwhile the turtle was outside the aquaterarium (according to the owner statement). It is not excluded that the turtle was taken from free-range, being already infected and then sold by the pet shop as a captive one.
In this reported case the lesions provoked by dog bite involved the plastron, the carapace, the liver and the lungs. It is known that deep, tearing wounds occur in dog bites and display perforations of shell corresponding to the intercanine distance. Hence, the distance between perforations were 55 mm on carapace and 46 mm on plastron, proving that the dog has bitten with the maxilar canines on carapace and mandibular ones on plastron.
Considering the findings gathered during the necropsy and the complementary exams, the cause of death was traumatic shock caused by the liver rupture and lung perforation. Concurrent lesions seem to be entirely the consequence of multiple dog bites.

CONCLUSIONS

A 1-year-old male of *Trachemys scripta elegans* presented numerous gross lesions featured by multiple fractures of the shell and skeleton induced by dog bite. The liver and the lungs were affected subsequently, death being induced by traumatic shock.
Spiroxys contortus infection was represented by gross and microscopic gastric injuries (ulcers and parasitary granulomas), this being the first case reported in captive turtle in *Trachemys scripta elegans.*

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