

Marine Turtle Newsletter

Case report: Lung Spiroorchidiasis in a Green Turtle (*Chelonia mydas*) in Southern Brazil

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Spiroorchiid trematodes are implicated as an important cause of stranding and mortality in sea turtles worldwide (Stacy *et al.* 2010). However, the real impact of these parasites on sea turtle health is poorly understood. The complete life cycle of marine spiroorchids still remains unknown; however snails or polychaete annelids may serve as intermediate hosts shedding cercariae that penetrate the mucous membranes of sea turtles, which are their final host (Dailey 1992). The adult trematodes inhabit the cardiovascular system, primarily the heart, as well as visceral and mesenteric vessels, where they copulate and oviposit, causing severe vasculitis, parasitic granulomas and thrombosis (Aguirre *et al.* 1998). Eggs may migrate and lodge in different tissues, where they induce a granulomatous response (Work *et al.* 2005). The disease is spread when infected turtles shed the parasite eggs in their feces or urine, through the cloaca (Dailey & Morris 1995).

Diagnosis of spiroorchidiasis in sea turtles is usually made at necropsy, when adult worms or eggs are observed either grossly or microscopically. The antemortem detection of spiroorchiid infection in wildlife is difficult, due to the common occurrence of subclinical infections, and is currently limited to serology (Work *et al.* 2005).

The blood flukes of turtles (Digenea: Spirorchiidae) and the blood flukes of crocodylians, birds and mammals (Digenea: Schistosomatidae) have long been considered as closely related, but distinct evolutionary lineages. However, recent morphological and molecular studies have considered these families as sister taxa within the Schistosomatoidea (Platt & Brooks 1997; Snyder 2004). The immune response to spiroorchid and schistosome eggs appears to be similar in their respective hosts. The arterial-dwelling spiroorchids release eggs in the direction of blood flow, resulting in a wide dissemination of eggs within the host (Platt & Brooks 1997).

On 28 January 2013, a juvenile green turtle (*Chelonia mydas*) was rescued by Projeto Tamar (Brazilian sea turtle conservation program) after stranding in São Francisco do Sul municipal district, in Santa Catarina State, Brazil. On admission, the animal measured 43.1 cm curved carapace length, 40.7 cm curved carapace width, and weighed 8 kg. The turtle was lethargic, weak and emaciated. Clinical signs included cachexia, anaemia (PCV 11%), dehydration, anorexia, abnormal respiratory sounds, increased respiratory rate and asymmetric floating, suggesting a true buoyancy problem. Death occurred a few days

after initial supportive care, and a necropsy was performed to determine the cause of death. During the procedure, visual examination of the gonads confirmed the turtle was female. All celomic organs were examined; however, gross changes were observed only in the lungs and consisted of multiple black nodules throughout the pulmonary parenchyma (Fig. 1).



Figure 1. Multiple black nodules occurring throughout the parenchyma of the lungs of a green turtle from São Francisco do Sul, Santa Catarina, Brazil.

Lung samples were collected and fixed in 10% neutral formalin solution and sent to the Laboratory of Animal Pathology, in northern Rio de Janeiro State University Darcy Ribeiro (UENF). The samples were cleaved and packaged in disposable plastic tissue cassettes. Infiltration and blocking were performed in paraffin, and leaf material was sliced into 5- μ m-thick sections using a rotary microtome. Sections were stained with hematoxylin and eosin (H&E) and mounted on the slide for subsequent histopathologic examination by light microscopy.

The histological analysis revealed lesions stemming from the presence of numerous fluke eggs, mostly located in the alveolar septum, where they frequently affect both airways and blood vessels. A chronic granulomatous pneumonia was characterized by heavy infiltrates of inflammatory cells, with multinucleated giant cells phagocytizing *Spirorchiid* eggs (Fig. 2).

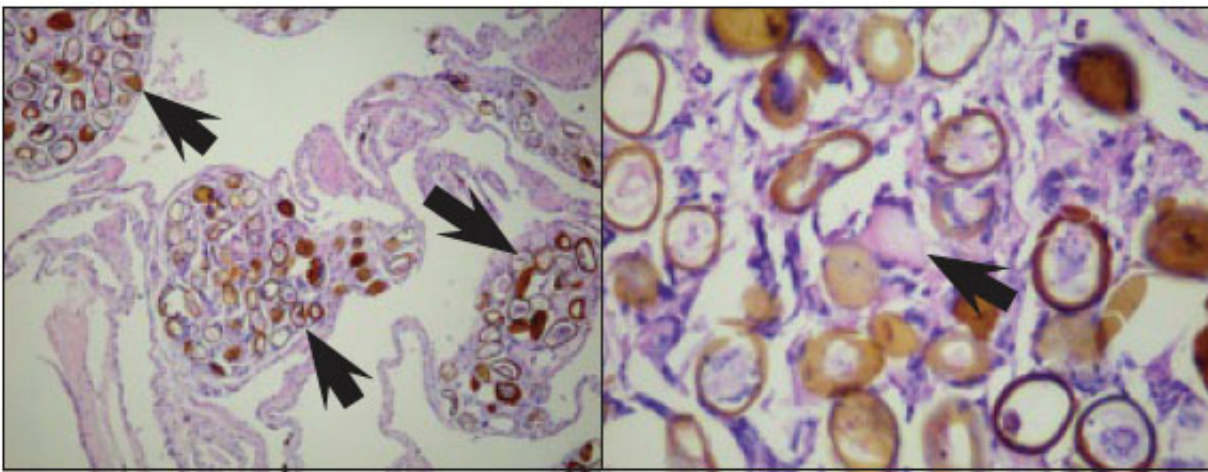


Figure 2. Representative photomicrographs of the lungs of a green turtle from São Francisco do Sul, Santa Catarina, Brazil. Left: Multiple eggs of *Spirorchis* spp. (arrows) with septal thickening and reduction of alveolar space. Parasite eggs being phagocytized by giant cells (H&E stain, X10 obj.). Right: Larger view of the previous image: Spirorchiid eggs being phagocytized by Langhans giant cells (arrow), resulting in a granulomatous pneumonia by Spirorchiid eggs (H&E stain, X20 obj.).

Spirorchidiasis is usually chronic and debilitating in its course, with most of the pathogenesis caused not directly by the adult worms but by the eggs they produce. The trematode eggs are released within the vascular system, reaching remote areas, such as the central nervous system (CNS) where they become lodged in small vessels, inciting a pronounced granulomatous reaction. The eggs may also migrate through blood vessel walls, causing damage and inflammation in adjacent tissues (Gordon *et al.* 1998). The most commonly affected tissues are the gastrointestinal tract, liver, spleen, lungs and CNS (Glazebrook *et al.* 1989). Clinical signs may include generalized debilitation, severe ulcerative colitis, pitted ulcerations (due to ischemic necrosis) of the carapace and plastron, edematous limbs due to vascular obstruction and buoyancy problems secondary to pneumonia (Norton 2005).

The most commonly reported therapy for Spirorchidiasis has been praziquantel, given orally at relatively high doses (50 mg/ kg) (Adnyana *et al.* 1997). Supportive care with fluid replacement and anti-inflammatory drugs may also be useful. It is relevant to cite that ALT and AST levels are expected to be elevated following treatment with this anthelmintic, due to dislodgement of dead flukes from mesenteric arteries and bile duct into the liver, causing release of hepatocellular enzymes into plasma (Adnyana *et al.* 1997). Unfortunately, although praziquantel is considered effective against adult trematodes, it may be only partly effective or ineffective against the parasite eggs (Norton 2005).

Detailed knowledge of parasite life cycles, with special reference to disease transmission and routes of infection, is essential to the understanding every aspect of host parasite interaction (Stacy *et al.* 2010), and may provide useful information regarding the implications for disease management.

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