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Death of a loggerhead sea turtle (*Caretta caretta*) from ingestion of an eel (*Myrichthys ocellatus*)

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Abstract: Gastrointestinal perforation in sea turtles may be associated with the ingestion of solid waste; however, other factors, including the ingestion of marine organisms, may cause intestinal perforation. Herein, we report, presumably, the first case of a death of a loggerhead sea turtle (*Caretta caretta*) from ingesting a live goldspotted eel (*Myrichthys ocellatus*) and describe the necropsy findings. The adult female loggerhead sea turtle was registered alive by a team of the Cetáceos da Costa Branca Project of Rio Grande do Norte State University (PCCB-UERN). The animal died in captivity after 8 days of an attempted rehabilitation process, and the carcass was immediately sent for necropsy. After incision, an encapsulated structure in the cranial region of the left lung was identified as a specimen of the ingested eel. The histopathological examination of the lung showed fibrin and numerous leukocytes, mainly macrophages, in the alveolus and bronchioles. The necropsy revealed that the ingestion of a live goldspotted eel (*M. ocellatus*) caused a gastric perforation in the turtle; this resulted in the displacement of the eel to the celomatic cavity with its cranial portion in the left lung, leading to sepsis and the consequent death of the turtle.

Keywords: Cheloniidae; gastric perforation; septicaemia; Testudines

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Loggerhead sea turtles (*C. caretta*) are a globally distributed migratory species and are classified as "vulnerable" to extinction according to the International Union for Conservation of Nature red list of threatened species (Casale and Tucker 2017). Conservation of this species is a complicated task that requires knowledge of its essential biological aspects, such as its reproduction, migration, and feeding habits (Burke et al. 1993).

These animals are carnivorous throughout their life cycle, preferentially feeding on fish, followed by pelagic tunicates, crustaceans, molluscs, and other invertebrates (Tomas et al. 2001; Revelles et al. 2007). The dietary data of these specimens are relevant to the understanding of their predator-prey relationships.

Moreover, it is difficult to diagnose gastrointestinal diseases in sea turtles antemortem. Clinical signs found on necropsies provide relevant decision-making information regarding the rehabilitation of stranded animals.

Herein, we report, presumably, the first case of a death of a loggerhead sea turtle (*C. caretta*) caused by ingesting a live goldspotted eel (*M. ocellatus*) and describe the necropsy findings.

Case description

An adult female loggerhead sea turtle (*C. caretta*), previously registered by the Cetáceos Costa Branca Project of Rio Grande do Norte State University (PCCB-UERN), was rescued alive on March 27, 2018 from Requenguela beach (4°39'56.4"S, 37°25'41.7"W), Icapuí, Ceará, Brazil.

The experiment was conducted with authorisation from the Ethics Committee on Animal Use (License No. 01/2019).

The prostrate and debilitated animal was rescued and sent to the Wild Animal Rehabilitation Center of the Cetáceos Costa Branca Project (UERN) in the city of Areia Branca, Rio Grande do Norte (RN), Brazil, where it was kept in a salt water enclosure measuring 200 cm in diameter and 75 cm in depth, with a water temperature of 25–28 °C and natural light from approximately 5:30 a.m. to 5:30 p.m. The biometric data included a curved carapace length of 96.0 cm, a curved carapace width of 87.0 cm, and a body weight of 91.0 kg.

The clinical examination revealed a regular body score, anorexia, noisy breathing, severe dehydration, and the presence of algae and sessile crustaceans (barnacles) on its carapace and plastron. The turtle received supportive treatment with fluid therapy (KabiPac®, 0.9% sodium chloride, 20 ml/kg i.v.; Fresenius Kabi, São Paulo, Brazil), diluted with a vitamin (Polivin B12®, vitamin B12, 0.3 ml/kg i.v.; Bravet, Rio de Janeiro, RJ, Brazil), antibiotic therapy (CEF 50®, ceftiofur hydrochloride, 4 mg/kg i.m.; Agener União, Embu-Guaçu, São Paulo, Brazil), and an analgesia (Tramal®, tramadol hydrochloride, 5 mg/kg s.c.; Grünenthal, Aachen, Germany; Maxicam®, meloxicam, 0.1 mg/kg i.m.; Ourofino, São Paulo, Brazil; D-500®, dipyrone, 25 mg/kg i.m.; Zoetis, Campinas, São Paulo, Brazil).

The turtle died after 8 days of treatment and was immediately sent for necropsy to the Marine Biota Monitoring Laboratory of UERN in Mossoró, RN.

A complete necropsy was performed following the standard *postmortem* protocol for sea turtles (Wyneken 2004). The celomatic cavity surrounding the entire plastron was opened for a macroscopic analysis and then removed for an *in situ* analysis of the organs. Subsequently, all the organs were removed outside of the cavity for evaluation, and the most evident macroscopic findings were photographed.

Organ fragments were collected for a histopathological examination and fixed in 10% formalin. After fixation for 48 h, the samples were dehydrated using increasing concentrations of alcohol (70%, 80%, 90%, 95%, 100%, 100%, and 100%) for a period of 1 h for each concentration, followed by diaphanization consisting of two xylol baths of 30 min each and a subsequent paraffin embedding by immersion in a granular histological paraffin with a melting point of 58–62 °C (Synth®; Labsynth, São Paulo, SP, Brazil) in an oven at 58 °C.

After preparation, the histological blocks were cut into 5-µm sections using a microtome (Leica RM2125 RT) adhered to glass slides, placed in an oven at 58 °C for deparaffinisation, and stained with haematoxylin and eosin (Prophet et al. 1992).

Photomicrographs of the most representative images were acquired under a light microscope (Leica ICC50 HD) and obtained using the Leica LAS EZ software (v2.0.0.; Leica Microsystems Ltd., Heerbrugg, Switzerland).

In the initial portion of the digestive tract, the oesophagus and stomach showed no food content, but a contained mucus; the cardiac region of the gastric mucosa showed a small, healed, rounded lesion

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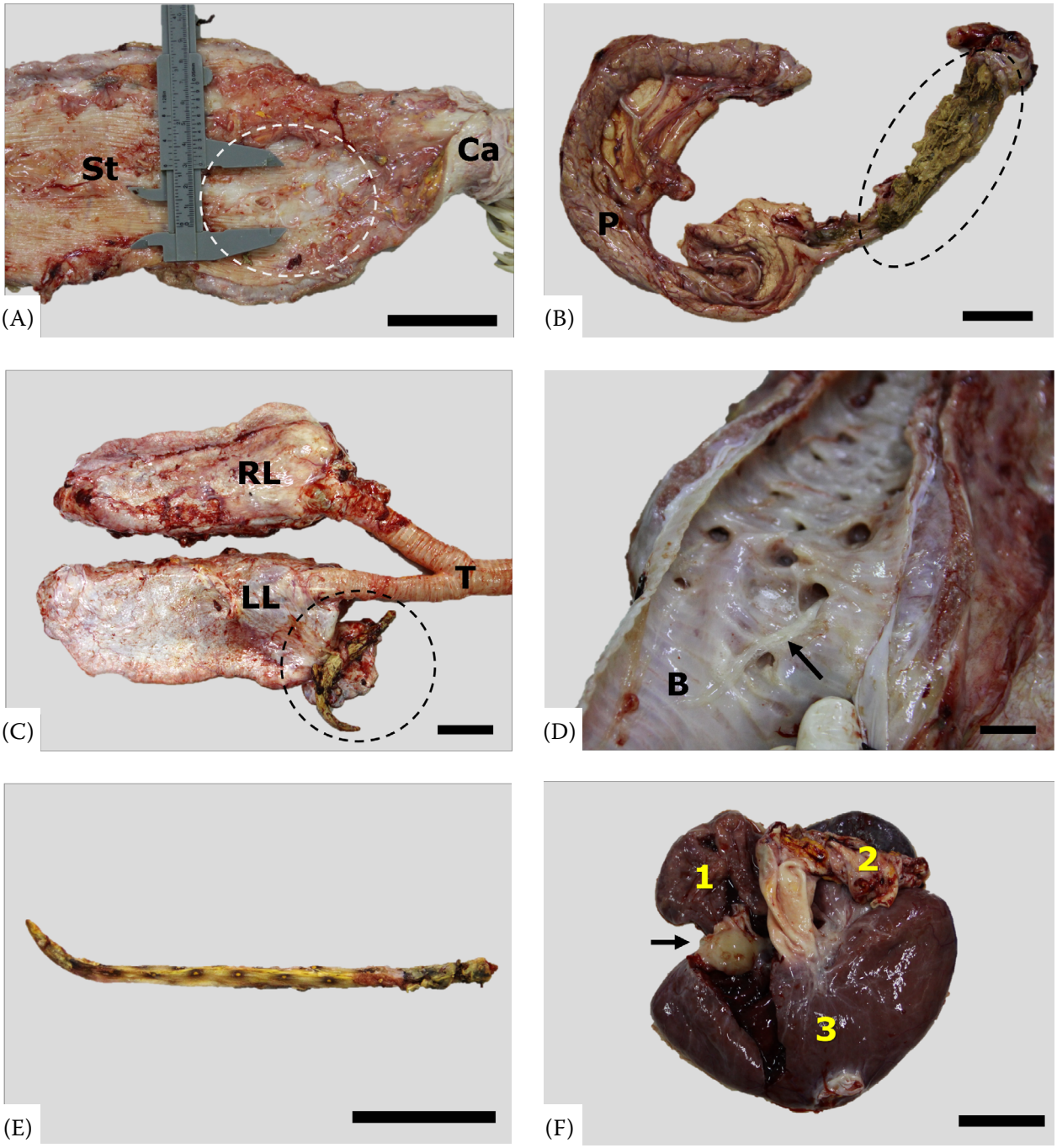

Figure 1. Macroscopic findings of a loggerhead seaturtle (*Caretta caretta*) that died from ingestion of a live goldspotted eel (*M. ocellatus*). (A) Perforation in the stomach (St), with presence of scar tissue (dashed circle) near the cardiac region (Ca) caused by the ingestion of the eel. Bar = 5.0 cm. (B) Presence of the encapsulated (dashed circle) extravasated stomach content in the pancreas (P). Bar = 5.0 cm. (C) Tracheal region (T) of the respiratory system and the right (RL) and left (LL) lungs with an *M. ocellatus* specimen in the cranial region of the left lung (LL). Bar = 5.0 cm. (D) Presence of a yellowish mucoid secretion in the bronchi (arrow). (E) Details of the *M. ocellatus* eel. Bar = 2.0 cm. (F) Dilated flaccid right atrium (1) with a thickened atrioventricular valve (arrow) and left atrium (2) and ventricle (3) without any apparent macroscopic changes. Bar = 5.0 cm

(Figure 1A). The intestines showed dilated portions containing gas and a greenish-brown liquid. The total length of the small and large intestines was 714 cm. An encapsulated foreign body was

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found adjacent to the pancreas, and an incision revealed that some unidentified food content extravasated from the stomach (Figure 1B).

The liver was enlarged, with rounded edges, capsular thickening, whitish granules distributed diffusely in the serosa, nutmeg-coloured parenchyma, and an abscess (10.0 cm diameter) with a caseous content (Figure 2A and 2B). The histopathological examination revealed multifocal areas of hepatocellular necrosis and hepatocyte cord dissociation (Figure 2C and 2D).

In the respiratory system, the trachea and bronchi showed a pale-yellow, mucoid secretion (Figure 1D) and solid parenchyma of firm and congestive consistency. An encapsulated structure in the cranial region of the left lung was identified as a specimen of the ingested eel after the incision. The eel was removed, and the specie was confirmed to be *M. ocellatus* (Figure 1C and 1E).

The histopathological examination of the lung showed fibrin and numerous leukocytes, mainly macrophages (Figure 3A and 3B), in the alveolus and bronchioles.

The tissue that surrounded the specimen in the left lung was predominantly vascularised fibrous tissue (Figure 3C and 3D).

In the heart, the right atrium was dilated and flaccid with a thickened atrioventricular valve. No apparent macroscopic changes were observed in the left atrium or ventricle (Figure 1F). The other organs showed no noteworthy macro- or microscopic changes.

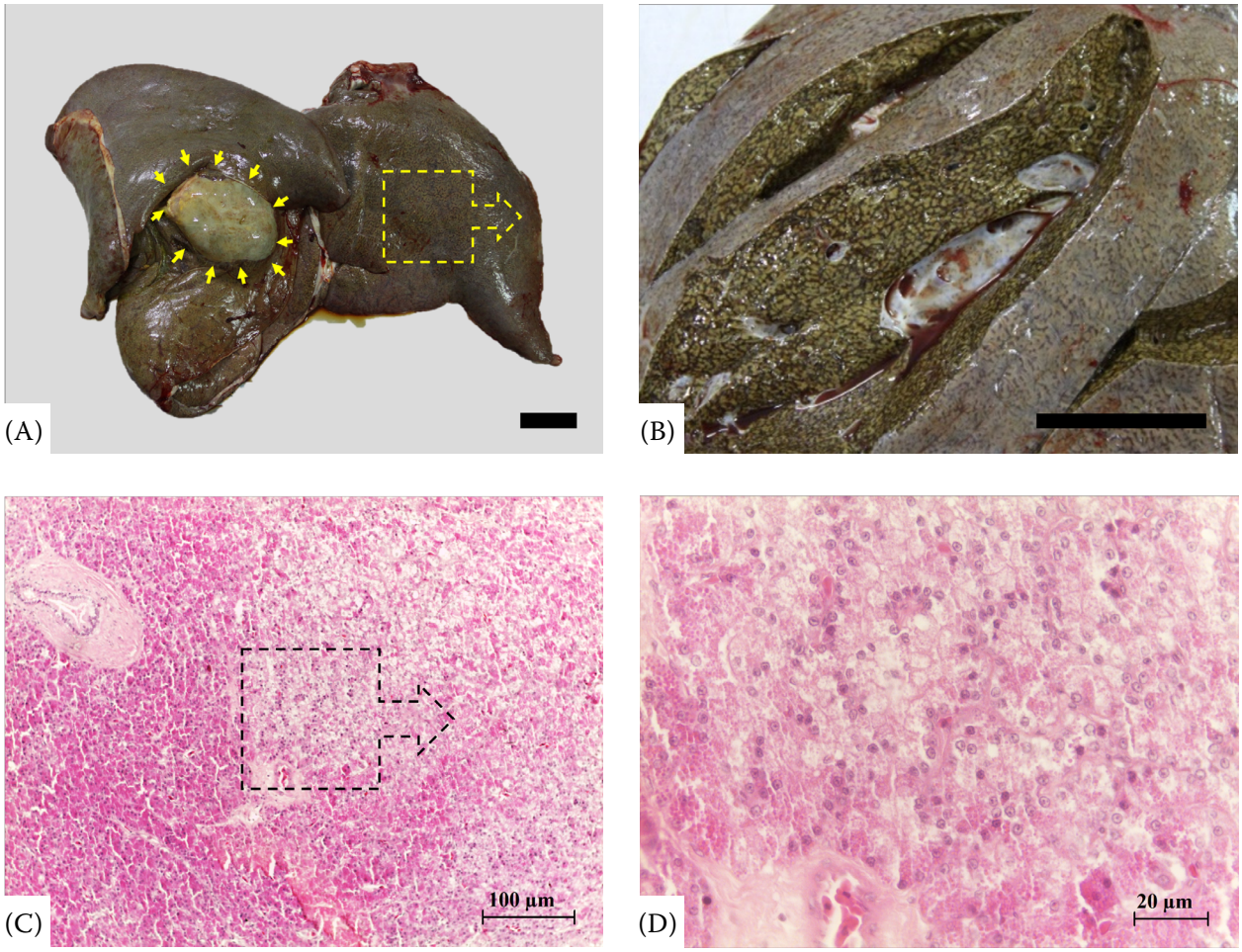

Figure 2. Histopathological findings of the liver of the loggerhead seaturtle (*Caretta caretta*) that died from ingesting a live eel (*M. ocellatus*). (A) Enlarged liver with rounded, thickened edges and a 10.0-cm diameter abscess with caseous content (arrows). Bar = 5.0 cm. (B) Nutmeg-coloured liver parenchyma. Bar = 5.0 cm. (C) Multifocal areas of hepatocellular necrosis. H&E; bar = 100 µm. (D) Zoomed Figure 2C showing the derangement of the cellular architecture resulting from hepatic necrosis. H&E; bar = 20 µm

H&E = haematoxylin and eosin

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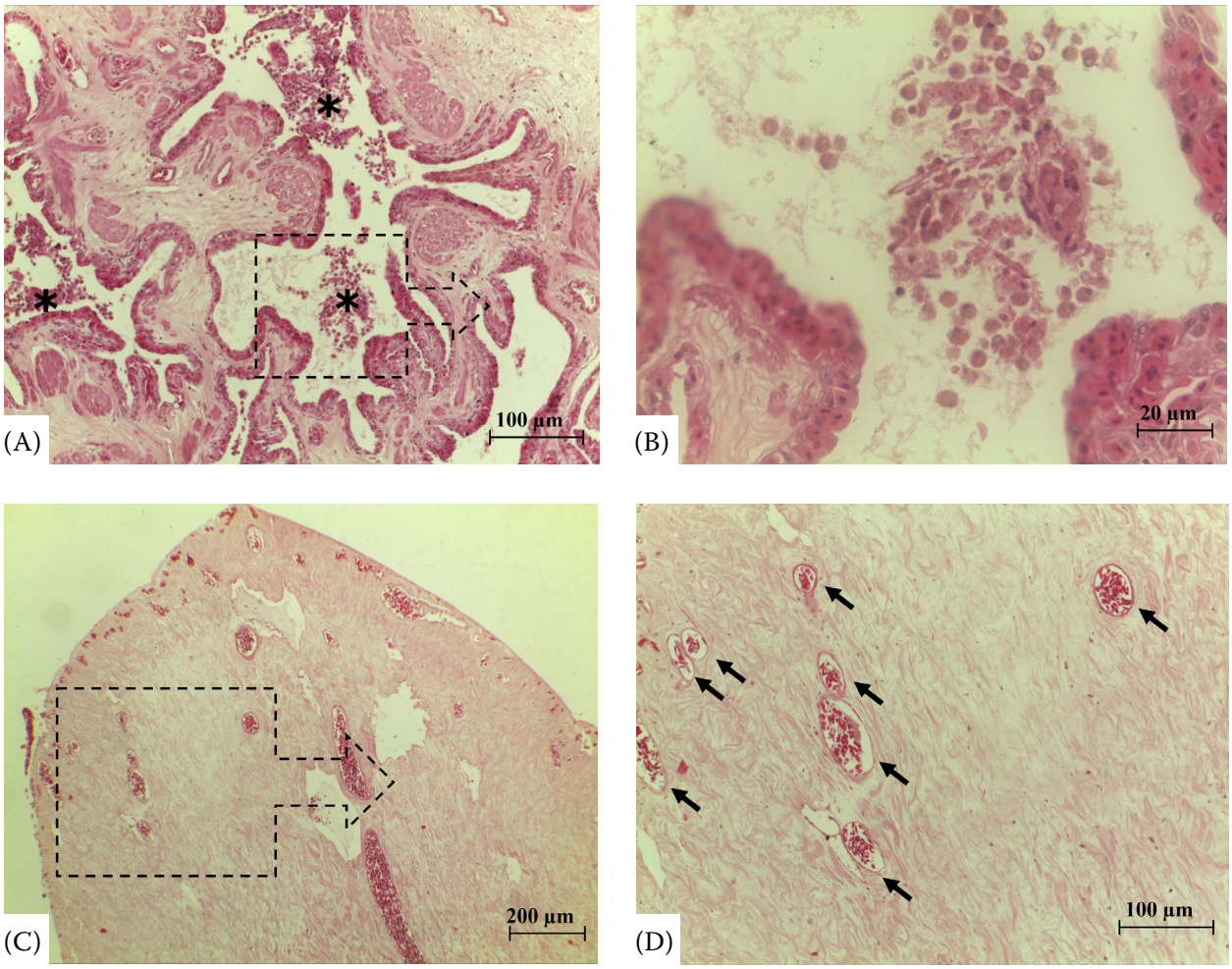

Figure 3. Histopathological findings of the lung of a loggerhead sea turtle lung (*C. caretta*) that died from ingesting a live eel (*M. ocellatus*). (A) Presence of an intra-bronchiolar fibrin-leukocyte exudate (*). H&E; bar = 100 µm. (B) Zoomed Figure 3A showing numerous macrophages and dendritic cells and abundant fibrin accumulation. H&E; bar = 20 µm. (C) Fibrovascular tissue that was involved in the eel specimen in the left lung. H&E; bar = 200 µm. (D) Zoomed Figure 3C showing a fibrous tissue with blood vessels (arrows). H&E; bar = 100 µm
H&E = haematoxylin and eosin

DISCUSSION AND CONCLUSIONS

Some studies on the feeding ecology of loggerhead sea turtles reported that the diet of these animals mainly consisted of fish, followed by pelagic tunicates, crustaceans, molluscs, and other invertebrates. Loggerhead sea turtles are often opportunistic feeders, mainly eating discarded fish; the prey quantity and diversity increases proportionally with the turtle size (Tomas et al. 2001; Revelles et al. 2007).

Goldspotted eels (*M. ocellatus*) are distributed throughout the Atlantic Ocean, from Bermuda to southern Brazil (McCosker et al. 2015). They are commonly found in north-eastern Brazil at depths of up to 30 m (Carvalho-Filho 1999). Despite being a prevalent species in the feeding areas of loggerhead sea turtles, any literature regarding eels being the feeding preference of these turtles or causing death is lacking.

In documented cases, gastrointestinal perforation in loggerhead turtles was associated with solid waste ingestion or the presence of hooks due to the incidental capture during fishing activities (Horau et al. 2014).

A study by Caracappa et al. (2018) on the incidental capture of *C. caretta* along the coast of Sicily reported that 129 of these loggerhead sea turtles had fishing hooks indifferent parts of the gastrointestinal tract, which were mostly found in the oesopha-

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gus (47.3%), intestine (24.8%), stomach (14.7%), and oral cavity (13.2%).

Mucosal lesions in the gastroenteric region lead to ischemia, necrosis, and perforation (Jerdy et al. 2017), possibly resulting in secondary bacterial infections and death, as in a case study reporting the bacterium *Enterococcus faecalis* in the urinary bladder, brain, intestine, kidney, liver, lung, and muscle of a loggerhead seaturtle with oesophageal perforation caused by a fishing hook and the intestinal intussusception caused by a nylon thread. In the present case, lesions in the oesophagus, intestine, or both were the probable routes of entry for this bacterium, and their presence in the organs and viscera suggested septicaemia as the cause of death (Fichi et al. 2016). The same mechanism could be underlying in the present case: The route of entry was the gastric perforation worsened by the presence of the eel in the lung. The necropsy findings in the lungs, such as an intra-bronchiolar fibrin-leukocyte exudate and cellular response, were consistent with a bacterial infection; other findings characteristic of septicaemia, such as hepatocellular necrosis and a thickened right atrioventricular valve (endocarditis) were also observed.

These findings revealed that the loggerhead sea turtle had ingested a live eel (*M. ocellatus*) that caused the gastric perforation, possibly displacing the eel into the celomatic cavity with its cranial portion in the left lung, and leading to sepsis and the consequent death of the turtle.

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Conflict of interest

The authors declare no conflict of interest.

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