

CASE STUDY

## Coccidiosis in an aquarium-managed common octopus (*Octopus vulgaris*): classical macroscopic and histologic features.

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Coccidiosis is the most prevalent disease in octopuses under human care; however, detailed macroscopic and histological examinations illustrating its hallmark features are scarce. This report presents a classic case of severe coccidiosis in an aquarium-derived formalin-fixed common octopus (*Octopus vulgaris*) as an educational resource for veterinarians working in aquatic medicine.

Octopuses play an integral role in the aquarium industry, with species such as the common octopus (*Octopus vulgaris*), giant Pacific octopus (*Enteroctopus dofleini*), curled octopus (*Eledone cirrhosa*), and California two-spot octopus (*Octopus bimaculoides*) frequently kept in aquaria and submitted to various specialist veterinary pathology services for examination (Rich et al. 2023). Despite the increasing popularity of these cephalopods, larger cohort pathology studies focused on octopuses under human care have only recently emerged, indicating the growing nature of this specialized field (Rich et al. 2023; Seeley et al. 2016; Fiddes et al. 2023; and Gestal et al. 2019). There remain few educational resources (e.g., gross images depicting key lesions) for pathologists, clinicians, and aquarists learning about cephalopods, in contrast to more traditionally studied aquatic species (e.g., salmonids, cyprinids) (Bruno, Noguera, and Poppe 2013; Roberts 2012).

Coccidiosis represents the most common infectious disease affecting octopuses under human care, with prevalence rates reaching as high as 50% in certain large aquarium cohorts (Rich et al. 2023) and 100% in some commercially wild-caught common octopus samples (Pascual et al. 1996). Despite its frequency in common octopuses, there is a scarcity of published gross and histological images for these infections, particularly when compared to the wealth of resources addressing coccidiosis in terrestrial species.

The paucity of pathologists performing necropsies on octopuses contributes to this deficiency, leading to specimens often being formalin-fixed either whole or as multiple tissues following basic dissection performed by aquarists to mitigate excessive autolysis (Andrew F. Rich, personal communication, November 2024). The submission of whole specimens allows pathologists

to gain invaluable insights into the complex anatomy of octopuses and provides an opportunity to capture images of classic lesions associated with coccidiosis and other diseases. This case study presents a typical example of coccidiosis in an aquarium-managed common octopus after formalin fixation with accompanying gross and histopathological images for reference and education in the field of veterinary pathology.

An adult (1-year-old), male, common octopus (*Octopus vulgaris*) with a history of poor appetite since its arrival at the housing UK aquarium (<1 year) was found dead in its tank unexpectedly. Prior to death, the individual had been housed alone in a tank following standard UK aquarium recommendations (i.e., 1000+ liter tank with an ideal minimum depth of 70 cm, an ideal temperature range of 15 to 20°C, low light levels with a 12-hour day/night cycle and a strong water flow rate [1.5x total volume per hour tank turnover] to maintain good water quality, to remove uneaten food, and to provide environmental stimulation) (BIAZA Aquarium Working Group 2024). Water quality parameters included: ammonia 0.05 mg/l [<0.00 mg/l], nitrite 0.02 mg/l [<0.00 mg/l], nitrate 35 mg/l [<50 mg/l], salinity 35.4 ppt [32-34 ppt], temperature 16.5°C [15-20°C], dissolved oxygen 96.9 [100], and pH 8.2 [around 8]. Its diet comprised a variety of crustaceans (e.g., small crabs), and mollusks (e.g. clams, whelks and mussels) based on availability.

The deceased octopus was removed from the tank and the cephalon partially incised to expose internal organs, before being fixed whole in a large volume of 10% neutral-buffered formalin-seawater solution (at the ratio of 1 part tissue to 10-part formalin solution) for 10 days, and subsequently submitted for gross inspection and histopathology post-fixation.

At gross inspection, the specimen weighed 524 g (post-fixation) and was rinsed using phosphate buffered saline (PBS) to reduce formalin vapor exposure prior to dissection in a class 2 biological safety cabinet. On external examination, segmentally extensive (18 cm length) skin ulceration was noted along the distal half of the L4 arm, revealing white eroded dermis. Small focal ulcers (up to 10 x 5 mm) were also noted near the tips of other arms. No mantle lesions were detected. On internal examination, an adult male reproductive tract was identified along with a reasonably sized digestive gland (liver equivalent and indicator of body condition) (Rich et al. 2023). Pathological lesions were primarily detected affecting the alimentary tract ([Figure 1](#)) characterized by numerous, off-white-to-cream, multifocal, firm, slightly raised, mucosal, intramural and subserosal hemispherical nodules (<1 mm diameter) affecting the esophagus, crop and stomach, with further diminishing involvement of the more distal segments (e.g., small intestine, caecum and large intestine). Similar nodules were also observed along the descending ducts of the secondary salivary gland. Renal appendages were also notably small/atrophic.

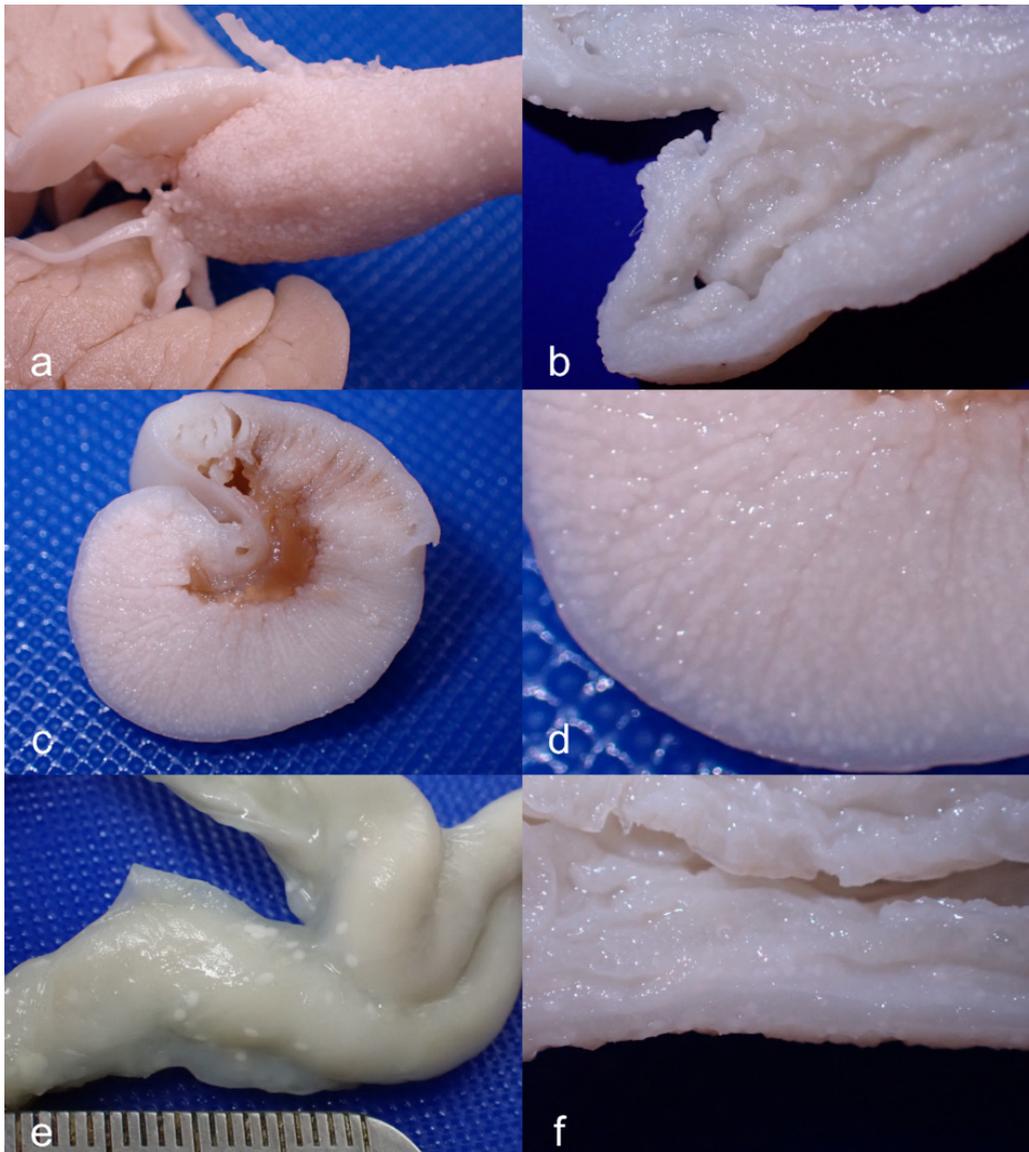


Figure 1. Coccidiosis in a formalin-fixed common octopus (*Octopus vulgaris*) – Gross pathology.

(a) Esophagus and crop – external surfaces. Numerous <1 mm diameter (miliary), slightly raised, off-white-to-cream, firm, subserosal nodules (coccidial oocysts) are widely scattered across the entire serosal surface. Posterior salivary glands are also present in this image. (b) Crop – cut internal surfaces. Similar <1 mm diameter, off-white-to-cream, discrete miliary nodules are present throughout the mucosal, intramural, and subserosal regions of the crop. (c) Cecum – cut internal surface. Characteristic innumerable page-like leaflets are seen. (d) Cecum – high power image of Fig 1C. Innumerable miliary nodules are present throughout the mucosal and intramural regions. (e) Distal intestine – Fewer yet moderate numbers of miliary nodules are seen widely scattered across the entire serosal surface. Scale intervals = 1mm. (f) Distal intestine – cut internal surfaces. Similar yet fewer, <1 mm diameter, off-white-to-cream, discrete miliary nodules are present throughout the mucosal, intramural, and subserosal regions of the intestine.

Histopathology revealed myriad coccidial oocysts throughout the alimentary tract, which mostly measured up to 600-800  $\mu\text{m}$  in size and were surrounded by thin fibrous walls and containing numerous internal sporocysts (each retaining between 4 to 8 sporozoites). These were embedded within the proprial connective tissues, muscle layers, and the subserosal connective tissues of the esophagus and crop ([Figures 2a and 2b](#)), plus the stomach ([Figure 2c](#)), cecum ([Figures 2d and 2e](#)), distal intestine ([Figure 2f](#)), and secondary salivary (venom) gland ducts to a lesser extent, with multifocal

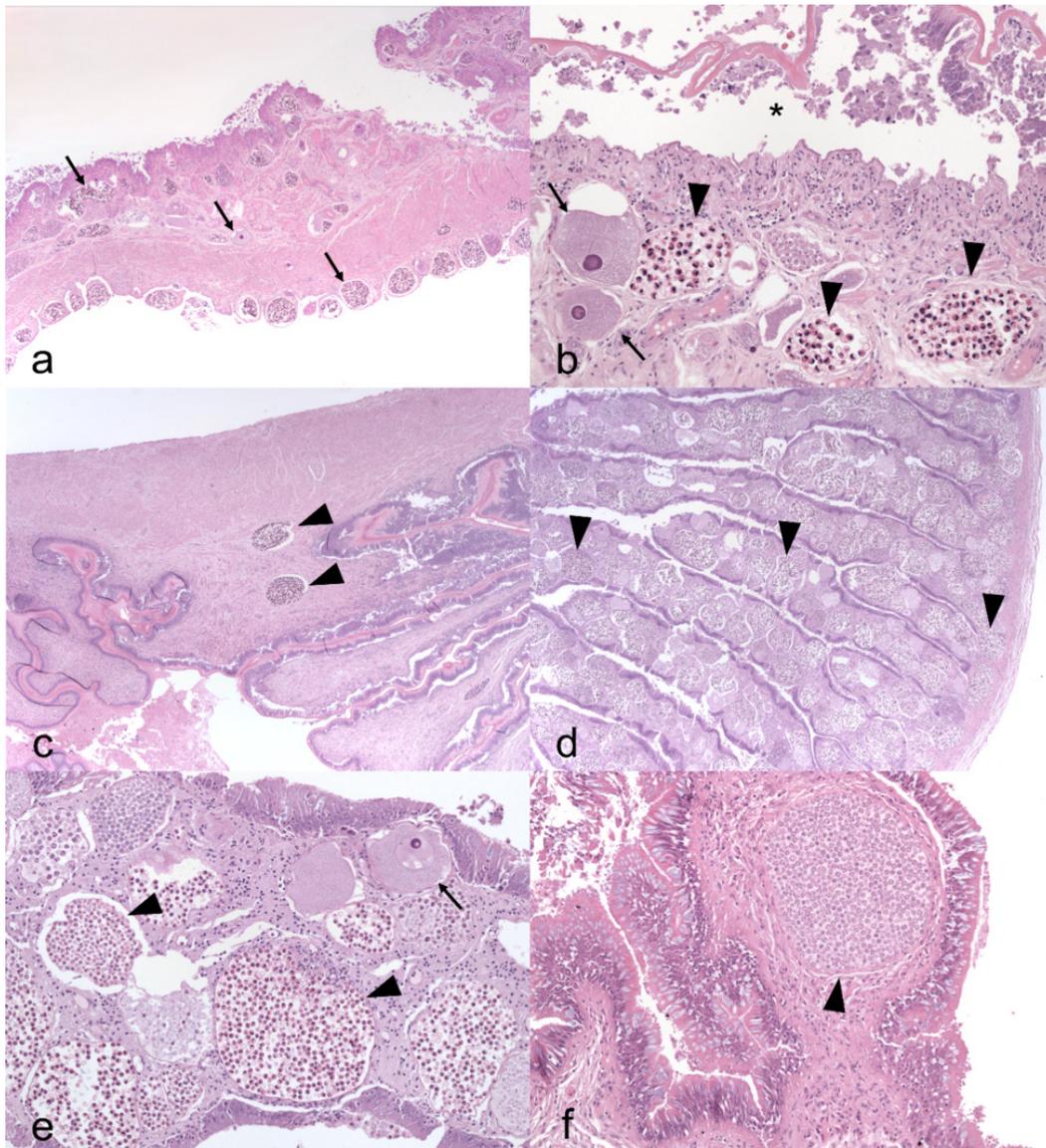


Figure 2. Coccidiosis in a common octopus (*Octopus vulgaris*) – Histopathology 1. Tissue distribution.

(a) Crop - Low power image demonstrating the widespread distribution of various coccidial stages (arrows) across the mucosa, muscle layers and subserosa. Hematoxylin & eosin (HE), 20x magnification (mag.). (b) Crop - Higher power image of Fig.2a. Subcuticular clefting (asterisk) with submucosal coccidial oocysts (closed arrowheads) and macrogametocytes (arrows) present. HE, 100x mag. (c) Stomach - Multiple coccidial oocysts (closed arrowheads) are present within the submucosa. HE, 20x mag. (d) Cecum - Numerous coccidial oocysts (closed arrowheads) are seen widely scattered throughout the lamina propria of the cecal leaflets. HE, 20x mag. (e) Cecum - Higher power image of Fig.2d. Submucosal coccidial oocysts (closed arrowheads) and occasional macrogametocytes (arrows) are present. HE, 100x mag. (f) Distal intestine - A coccidial oocyst (closed arrowhead) within the intestinal lamina propria. HE, 100x mag.

areas of mixed inflammatory infiltrates (large granulocytes and smaller granulocytes (synonym: hemocytes)) (Castellanos-Martínez et al. 2014) where oocysts had ruptured.

Alongside coccidial oocysts, occasional intralesional microgametocytes and macrogametocytes were also present (Figure 3). Segmentally, there was mild to marked disruption of the alimentary mucosa with superficial cuticle necrosis, mucosal ulceration and secondary liberation of coccidia and inflammatory cells into the alimentary tract lumen. Species identification

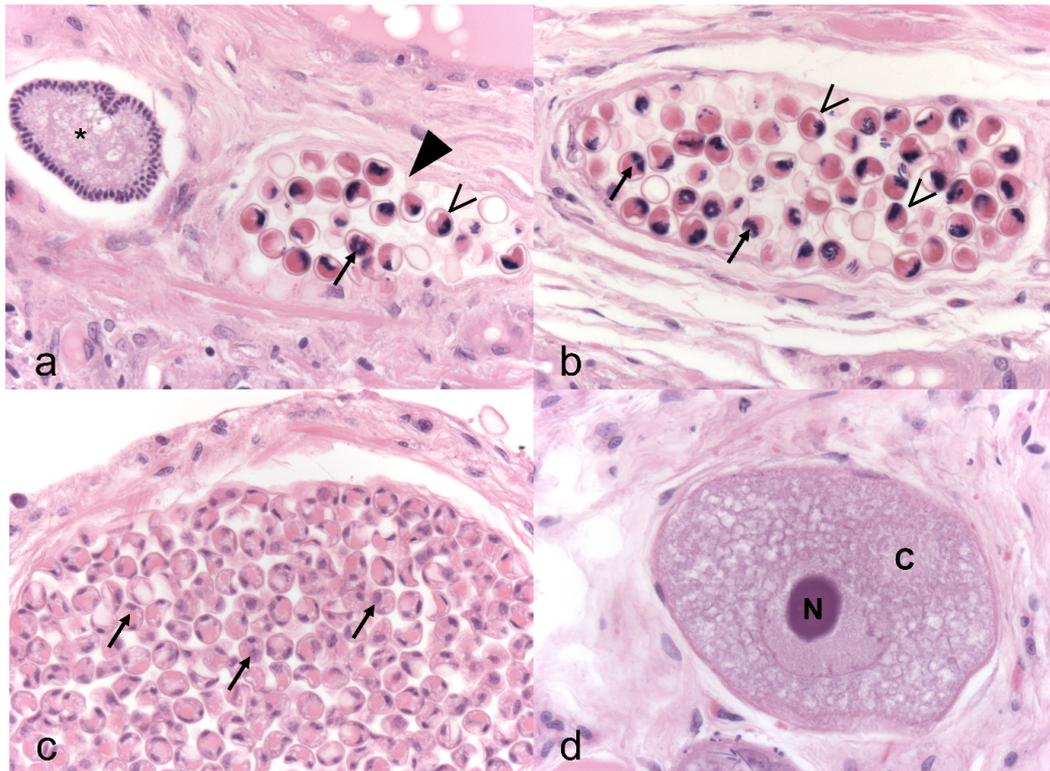


Figure 3. Coccidiosis in a common octopus (*Octopus vulgaris*) – Histopathology 2. Coccidial morphology.

(a) Coccidial microgametocyte (asterisk) and a small coccidial oocyst (closed arrowhead), the latter retaining multiple internal sporocysts (open arrowhead) (each retaining between 4 to 8 sporozoites (arrow)). HE, 400x mag. (b) Coccidial oocyst, approximately 800µm greatest diameter, containing numerous sporocysts (open arrowhead) retaining sporozoites (arrows). HE, 100x mag. (c) Coccidial oocyst demonstrating another developmental sporozoite stage (arrows). HE, 400x mag. (d) Coccidial macrogametocyte with a single distinctive nucleus (“N”) and foamy cytoplasm (“C”). HE, 400x mag.

of coccidial species requires molecular analysis and electron microscopy, techniques beyond the diagnostic scope and finances at the time of the investigation. *Aggregata octopiana* is favored in this common octopus, based on previously documented host-parasite associations and histomorphology (Gestal, Abollo, and Pascual 2002; Gestal et al. 2002; Castellanos-Martínez et al. 2019). Concurrent histological findings included 1) severe, diffuse, active-on-chronic renal tubular degeneration with interstitial fibrosis (chronic kidney disease) and small granulocytic inflammation, 2) moderate, multifocal, ulcerative and mixed granulocytic dermatitis of the arms, 3) mild, diffuse, small granulocytic branchitis with mild multifocal colonization by epitheliocystis-like organisms, 4) disseminated vascular mixed granulocytosis, and 5) mild, multifocal-to-coalescing, small granulocytic orchitis.

Intracellular coccidian parasites that infect the coleoidea subclass of cephalopods (i.e., octopus, cuttlefish, and squid) are primarily of the genus *Aggregata*, with *A. octopiana* being most significant in *O. vulgaris* and with widespread geographical distribution (Gestal et al. 2002; Gestal, Abollo, and Pascual 2002; Castellanos-Martínez et al. 2019; Tedesco et al. 2017; Castellanos-Martínez, Pérez-Losada, and Gestal 2013; Betancor et al. 2013). *Aggregata* spp. exhibit two-host life cycles with gametogony and sporogony occurring in the alimentary tract of the cephalopod definitive host, causing

mucosal ulceration and sloughing of mixed parasitized necrotic cellular debris, thereby allowing the release of new infective stages (i.e., sporocysts containing infective sporozoites) into the feces. This material is ingested by a crustacean intermediate host (e.g., crab), allowing schizogony and the development of merozoites. These merozoites subsequently infect the cephalopod when the intermediate host is consumed (Gestal et al. 2002; Gestal, Abollo, and Pascual 2002; Castellanos-Martínez et al. 2019; Tedesco et al. 2017). The significance of this life cycle to aquaria housing cephalopods is that infected octopuses are unlikely to transmit infection to other cephalopods within exhibits.

Although coccidial infections represent a pathogenic risk to captive cephalopods, a recent large-cohort review of pathological findings in cephalopods under laboratory and aquarium management suggested that coccidiosis in different octopus species varies in its pathogenicity (Rich et al. 2023). Specifically, whilst common octopuses and giant Pacific octopuses were more likely to be infected with coccidia (prevalences of 68.4% and 46.3% respectively in that study), this was judged to be the cause of death in only 32.3% and 36.0% of such cases. In contrast, despite a lower prevalence (23.8%) in California two-spot octopuses, such infections were considered a potential cause of death in 60% of cases. In the common octopus described here, the abundance of coccidial parasites and evidence of substantial enteric pathology suggests coccidiosis was a contributor to the clinical presentation and death. *A. octopiana* is the most likely coccidian species in this case, based on established host–parasite associations and characteristic histomorphology. However, future confirmation using molecular or ultrastructural techniques, such as electron microscopy, next-generation sequencing, or broader commercial availability of the recently developed quantitative real-time PCR assay for *A. octopiana* (Pérez-Polo et al. 2025) would be invaluable for definitive species identification in captive octopuses. These approaches may also help determine whether histomorphologically similar but pathogenically distinct *Aggregata* species contribute to the observed variability in disease severity in *Octopus vulgaris*. Concurrent systemic inflammation characterized by vascular granulocytosis and small granulocytic infiltrates in other organ systems indicated a more widespread inflammatory response to which coccidial-induced enteric mucosal injury and concurrent dermatitis lesions were likely contributory, perhaps through the development of bacterial sepsis. Bacteria were not detected histologically but microbial cultures could not be performed due to the exposure of the specimen to formalin prior to gross inspection. Severe chronic renal disease was a further contributing factor in the death of this animal.

In summary, this case represents a classic example of coccidiosis in a common octopus and given the severity of the infection within the proximal alimentary tract, it is likely that coccidiosis may have contributed to the reduced appetite documented in this animal's clinical history.

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This article is dedicated to the memory of Stephen Andrew Warren Rich (11.12.1953 – 09.07.2024) – a beloved father and skilled canoeist who inspired his son’s fascination of the aquatic world and shared a love of octopuses.

### ***AUTHOR CONTRIBUTIONS***

AFR and ILP both examined the formalin-fixed specimen macroscopically, photographed the lesions, performed background research on the topic, conceived/designed the project, and wrote the manuscript. AFR examined the specimen histologically. MFS reviewed the results and edited the manuscript.

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