



Case report

FIRST REPORT OF ASPERGILLOSIS IN A FREE-RANGING GREAT CORMORANT (*PHALACROCORAX CARBO*, LINNAEUS, 1758)

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Summary

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This report describes the first case of aspergillosis by *Aspergillus fumigatus* in a free-ranging great cormorant (*Phalacrocorax carbo*, Linnaeus, 1758) in Northern Portugal. The disease was observed after death and the agent was confirmed with histology and microbiology examinations. It is possible that the animal was immunosuppressed from a previous disease or due to the accumulation of pollutants in the tissues. This report suggests that aspergillosis in wild birds could be more prevalent than previously thought and highlights the importance of post-mortem examinations in wildlife.

Key words: aquatic birds, aspergillosis, fungi, *Phalacrocorax carbo*

The great cormorant (*Phalacrocorax carbo*, Linnaeus, 1758) is an aquatic bird from the Order Suliformes, Family *Phalacrocoracidae* (Liordos, 2004). These animals can be found throughout Europe, Asia, Africa, Australia, and northeastern coastal North America (Schaub & Kéry, 2022). This species is classified as Least Concerned (LC) according to the IUCN Red List of Threatened Animals (IUCN, 2018). Their main threats are hunting and poisoning (persecution by the aquaculture industry), displacement from offshore renewable energy, bycatch, oil spills, viral

diseases, climate change, recreational activities (e.g. aquatic sports) and pollution (Dewey, 2024).

Aspergillosis is a fungal infection in birds, caused by the *Aspergillus* species, particularly *Aspergillus fumigatus* (Arné *et al.*, 2021). This infection primarily affects the respiratory system, leading to symptoms such as difficulty breathing, nasal discharge, and lethargy. Birds with compromised immune systems, those under stress, or those living in unsanitary conditions are more susceptible (Beer-naert *et al.*, 2010). The disease can be

manifested in acute or chronic forms; acute cases often result in rapid decline and death, while chronic cases may show prolonged respiratory issues (Beernaert *et al.*, 2010; Melo *et al.*, 2020a). Diagnosis typically involves clinical signs, radiography, endoscopy, and fungal culture or PCR testing (Fischer & Lierz, 2015). Treatment includes antifungal medications, supportive care, and environmental management to reduce fungal spore exposure. Prevention focuses on maintaining clean living conditions, reducing stress, and ensuring a healthy diet to bolster the bird's immune system (Beernaert *et al.*, 2010; Melo *et al.*, 2020b).

Case description

An adult male great cormorant (*Phalacrocorax carbo*, Linnaeus, 1758) was admitted

to wildlife rehabilitation centre in Northern Portugal after being found debilitated. The clinical examination demonstrated a poor body condition (2 on a scale of 0 to 5), dehydration (10%) and small lacerations on the beak and face. Support treatment was provided, but the animal eventually died a few days later. A *post-mortem* examination revealed that the animal presented dehydration, poor body condition, and pallid mucosae (Fig. 1A). Internal examination evidenced that the right air sacs (abdominal, posterior thoracic, and anterior thoracic) were thickened and yellowish-white in colour (Fig. 1B). In the caudal portion of the right lung, whitish to yellowish nodules with a firm texture upon palpation were observed. Areas of inflammation surrounded the lesions, and there was caseous

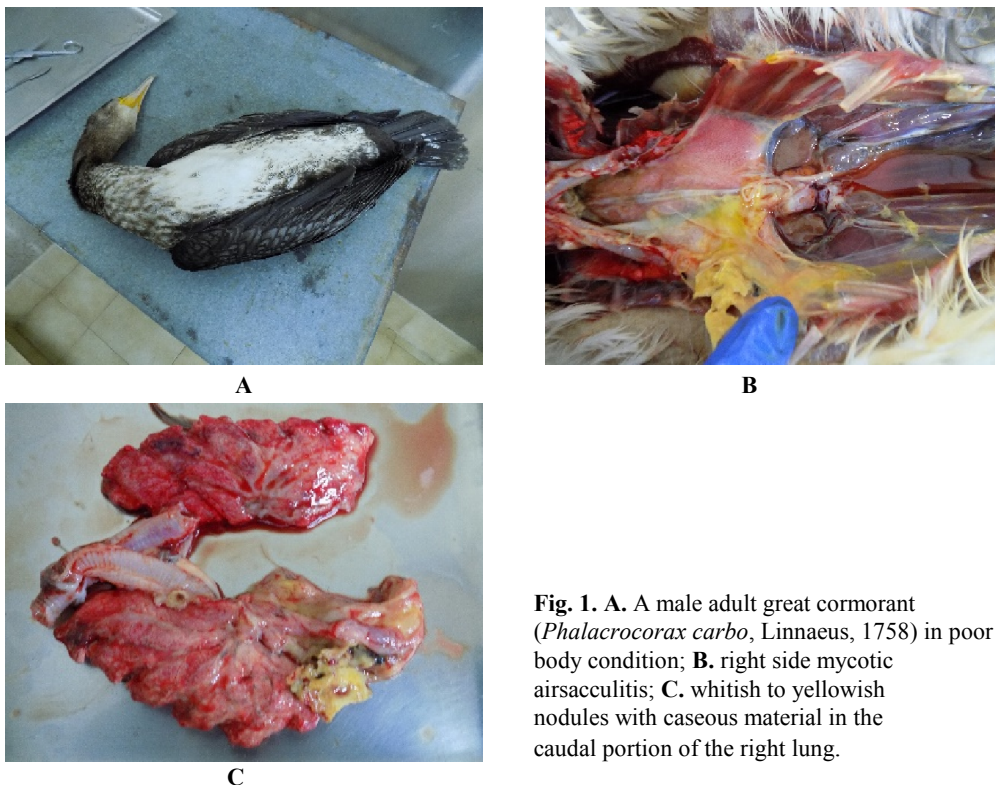


Fig. 1. A. A male adult great cormorant (*Phalacrocorax carbo*, Linnaeus, 1758) in poor body condition; B. right side mycotic airsacculitis; C. whitish to yellowish nodules with caseous material in the caudal portion of the right lung.

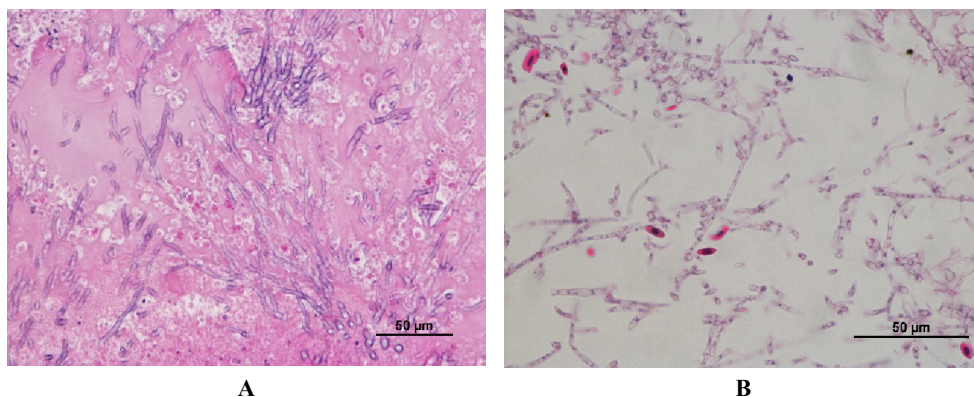


Fig. 2. A. Histological cross-section exhibiting invasive pulmonary *Aspergillus* spp. (aspergillosis) with septated hyphae and acute-angled branching (H&E, scale bar=40 µm, 10× magnification); **B.** Septate hyphae, forms with conidial heads (aspergillosis) (scale bar=50 µm, 40× magnification).

(cheese-like) material within the nodules (Fig. 1C). The other organs did not present any lesions.

Samples of air sacs and lungs were collected for histopathology and stained with haematoxylin-eosin and Grocott. Multifocal to coalescing heterophilic granulomas displaying necrosis of heterophils, and giant multinucleated cells were observed. Thin (3–12 µm), non-pigmented (hyaline), septated hyphae with homogenous acute angle branching consistent with *Aspergillus* spp. invading the granulomas, lung surface and air sacs were present (Fig. 2 A,B). The final diagnosis was mycotic airsacculitis and pneumonia.

Portions of injured air sacs and lungs, liver and kidney were aseptically collected and seeded onto Sabouraud Dextrose Agar (BioMérieux, France) for mycological culture at 25 °C. On the fungal plates, fungal colonies developed with olive to dark green colour on the upper surface, and cream reverse consisting of a dense felt of erect conidiophores (Fig. 3A). The microscopic examination performed using Lactophenol Cotton Blue

(Merck, Germany) staining showed aspergilla with columnar conidial heads. The fungus was identified as *Aspergillus fumigatus* based on its macroscopic and microscopic morphological characteristics (Fig. 3B).

Aspergillosis has been reported in numerous species of birds (Melo *et al.*, 2020b; Arné *et al.*, 2021). To the authors' knowledge, this disease has been described in great cormorant (*Phalacrocorax carbo*) for the first time (Garcês *et al.*, 2018). There are reports of aspergillosis in other cormorant species, such as the neotropical cormorant (*Nannopterum brasiliense*) (Melo *et al.*, 2020a), double-crested cormorants (*Phalacrocorax auritus*) (Bicknell *et al.*, 1971) and Japanese cormorant (*Phalacrocorax capillatus*) (Lim *et al.*, 2016).

Aspergillosis is associated with high mortality in animals in captivity kept in zoological institutions, sanctuaries or wildlife rehabilitation centres. Information regarding free-ranging wild animals is very scarce (Melo *et al.*, 2020a). Nowadays, the period of captivity in wildlife rehabilitation centres is too short (a few

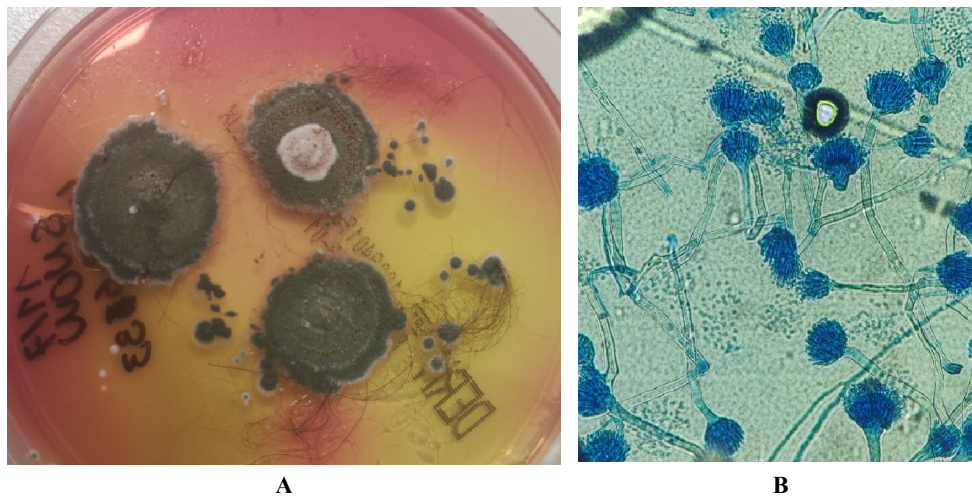


Fig. 3. A. Fungal colonies with olive to dark green colour on the upper surface, and cream reverse consisting of a dense felt of erect conidiophores on Sabouraud Dextrose Agar; **B.** *Aspergilla* with columnar conidial heads compatible with *Aspergillus fumigatus* stained with lactophenol cotton blue (40× magnification).

days) to develop this pathology, so most likely the animal became infected with the fungus in the wild. The source of contamination is unknown. *Aspergillus fumigatus* is a ubiquitous organism as many birds are carriers of its spores in the lungs and air sacs, collected from rotting or decaying vegetable material (Nardoni *et al.*, 2006). It is possible that the animal was immunosuppressed from a previous disease or from accumulation of pollutants in the tissues. This report suggests that aspergillosis in wild birds could be more prevalent than previously thought and highlights the importance of *post mortem* examinations in wildlife.

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