DIAGNOSING THE FUZZIES

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Mycotic infections can be divided into two basic types based on the fungal phase that the organism is most commonly observed—the mycelial phase and the yeast phase. The mycelial phase most commonly observed is *Aspergillus* and most common species is *fumigatus*. It is this mycelial phase that is the "fuzzie." However, this article will focus on several fungal pathogens in birds—*Aspergillus* sp, *Candidia* sp, and *Macrorhabdus ornithogaster*—and how to diagnose these infections.

ASPERGILLOSIS

Pathogenesis

One of the most common or at least most difficult-to-treat respiratory infections is from *Aspergillus* sp. Free-living birds tend to be free of *Aspergillus* species, and serologic testing of antibodies in raptors has been negative. Turkeys infected with *Aspergillus* produce moderate to severe fibrino-heterophilic or granulomatous air sacculitis. The air sacs appeared thickened, opaque, and or filled with fibrinous to gelatinous material. Macrophages invade the area quickly along with heterophils. Edema and fibroplasia develop early on, which stiffens the air sac, reducing its functional capabilities. Around 7 days into the disease process, lymphoid hyperphasia develops. It is important to note that titer values (obtained from AGID and ELISA tests) were not helpful clinically at predicting mortality—only weight loss was helpful in these research studies using turkeys.

There is also concern that bacterial components including the lipopolysaccharide (LPS) portion of the cell wall in *Pasteurella mutocida* reduces pulmonary performance initially, setting up an *Aspergillus* infection. LPS may also interfere with the phagocytic role of the epithelial lining of the lung and air sacs. Gliotoxins produced by some strains of *Aspergillus* sp may be an important factor in the pathophysiology of aspergillosis, with direct cytotoxic effects and immunosuppression.

Diagnosis

Aspergillosis can be difficult to diagnose in companion psittacine species. These birds can present with a chronic or acute infection. The acute form results in clinical signs in a few days: dyspnea, lethargy, depression, anorexia, weight loss, emaciation, exaggerated respiratory effort, cyanosis, and sudden death. The chronic form is often referred to with respiratory tract signs through an immunocompromising event. Signs include a change in voice character or quality, loss of voice, respiratory stridor, ataxia, torticollis, and possible seizures.

No definitive test is available, producing black or white results. Instead the clinician must rely on multiple tests and experience in arriving at a diagnosis. Hematologic findings can include a heterophilic leucocytosis, monocytosis, lymphopenia, hyper-proteinemia, and a nonregenerative anemia. The total WBC is often > 20,000/μL but may be in the normal range when the bird experiences immune suppression. The plasma biochemical analytes are nonspecific with AST and CK commonly elevated, but not consistently. Radiographs do not typically demonstrate visible lesions until the latter stages of disease when the patient decompensates. Radiographic observations include a prominent parabronchial pattern, loss of definition or asymmetry of the air sacs, hyperinflation of the abdominal air sacs, thickening of the air sac membranes, and soft tissue opacities within the respiratory tree.

Endoscopy allows for visual inspection of the respiratory tree and biopsy for cytology and culture. White, yellow, grey, or green plaques can be visualized lining the walls of the air sacs and within the lung parenchyma during the latter stages of the disease process.

The swab or biopsy should be cultured on Sabourad's dextrose agar. Culture of the organism in the absence of lesions, however, is not diagnostic. However, Gram stains and culture can help to identify the organisms obtained from the plaque. Usually, *Aspergillus* colonies form blue-green colonies at 48 hours.

Plasma or serum electrophoresis can be useful when monitoring disease progression and the response to therapy. Acute infections often demonstrate an increase in beta globulins, while chronic ones show an increase in beta or gamma fractions or both. However, birds with immune suppression may have hypoproteinemia.

There are several labs with ELISA tests developed for the detection and levels of *Aspergillus* antibody. These tests tend to perform better with chronically infected birds except for owls. However, false positives have been noted as well as false negatives, particularly with immune-suppressed birds or those with relatively walled-off granulomas. One lab provides an ELISA test of the detection of antigen and was adapted from human use. This test also requires clinical experience for interpretation as false positives and negatives occur.

CANDIDIASIS

Candida has been identified as an increasingly prevalent infectious agent in a variety of animals and has caused severe disease in humans. Gram-negative bacteria have been replaced with gram-positive bacteria and *Candida* as the most common bloodstream pathogens in humans in hospitals in the United States. Previously considered opportunistic diseases, particularly in neonates, they are increasingly difficult to cure. Normally the host defense system and the normal bacterial flora of the gastrointestinal tract keep yeasts like *Candida* under control. Although small numbers of non-budding organisms are considered normal flora in healthy florivores, in this author's experience, they are not present except when carried into the tract by food items, such as breads.

Candida affects the mucocutaneous surfaces that often include the GI tract and in particular the oropharynx, crop, and esophagus. Common signs included delayed crop emptying, crop stasis, regurgitation, anorexia, depression, and poor digestion of food. In chicks where the upper GI tract is affected, they do not grow properly and may appear stunted. The droppings of birds with GI tract involvement are abnormal and may be watery. Cockatiels, budgerigars, and cockatoos are considered higher risk psittacine species. Systemic infection has resulted in CNS signs in a flock of canaries, along with GI signs.

Candidiasis, when it affects the crop may be called thrush in raptors or sour crop in psittacines. It is most commonly caused from *Candida albicans* but may be from *C. parapsilosis*, *C. krusei*, and *C. tropicalis*. In young birds it is associated with improper handfeeding and a noncompetent immune system. These infections can often result from the administration of antimicrobials in adult and young birds. Antifungals specific for yeasts may need to be considered in the treatment plan of the avian patient.

The diagnosis of candidiasis may be difficult if the lesions are deep within the GI tract. Gram stains of swabs or biopsy specimens are important for identifying gram-positive budding yeasts or those with pseudohyphae. Flushes using pH balanced solutions of the nares and upper respiratory tract are helpful and examination are helpful in determining if these organisms are involved with the infection or complicating it. *Candida albicans* are basophilic gram positive oval shaped organisms measuring 3.5–6.0 x 6–10 µm. The presence of pseudohyphae suggests a more severe infection. Systemic candidiasis is currently rare in birds but may become more common, as it is in humans. In these cases, *Candida* sp. may be isolated from the blood, bone marrow, and/or parenchymatous organs.

MACRORHABDIASIS

Macrorhabdus ornithogaster has only been recently identified as a yeast, as it was previously thought to be a large rod-shaped bacteria, a "megabacterium." This organism was implicated in a syndrome of budgerigars that was described as "going light." It is a long straight rod 3 to 4 µm wide and 20 to 80 µm long. The organism may be longer as they often occur as long chains of cells where the separation between them are not visualized. On a gram stain, the thick cell wall may not stain with only the cytoplasm staining and they may not take up common cytology stains. For this reason, it is often difficult to visualize this organism, particularly the live bird.

Proventricular scrapings or a flush from the proventriculus is used for an antemortem diagnosis. Sick birds are more likely to shed organisms in their droppings. Wet mounts of unstained droppings examined under 40 and 100x and with a lowered diaphragm are more likely to be rewarding. Often multiple samples will need to be examined to find the organism. A new PCR test may be helpful at diagnosing this organism.

The implication of *Macrorhadus ornithogaster* producing disease remains unclear. Birds may shed organisms but appear normal on physical examination. Sick birds may shed a large number of organisms but may not shed any. It may be considered a pathogen while others believe it to be a commensal.

The organism appears to be prevalent in budgerigar aviaries with fecal shedding ranging from 27% to 64% based on fecal shedding. It has also been commonly observed in parrotlets, cockatiels, and lovebirds. Additionally the organism has been found in chickens, turkeys, geese, ducks, and ostriches.

In budgerigars, there may be an acute or a chronic presentation. Birds presenting with an acute presentation suddenly go off feed and regurgitate with or without bloodstaining. Death occurs within several days. The chronic form may be more common. Budgies appear hungry and are observed grinding food in their beak but not ingesting it. Regurgitation is common including birds presenting with matted feathers with regurgitated material around their heads. Undigested seeds may be found in the droppings. Other birds may have diarrhea with or without melena. These birds will have reduced pectoral muscle mass and appear to be "going light." These signs are not pathognomonic for this disease as candidiasis and other conditions may present similarly.

Parrotlets and lovebirds present with similar signs as with budgies. Parrotlets were observed to present acutely with regurgitation and melena. Lovebirds were observed to have significant numbers of organisms in their droppings but were also infected with pscittacine beak and feather disease (PBFD).

RECOMMENDED READING

1. Orosz SE. Antifungal drug therapy in avian species. Vet Clin North Am Exotic Anim Pract. 2003;6(2):337-350.
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