



Diagnostic Exercise

From The Davis-Thompson Foundation*



Case #: **159**, Month: **February**, Year: **2021**

Answer Sheet

Title: *Mycotic periorchid granuloma*

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Clinical History: A 2-year-old Egyptian Fayoumi rooster had sudden onset of limping in the right leg prior to rapidly collapsing.

Necropsy Findings:

There was blood in the oral cavity along with 85 ml of clotted blood in the coelom.

Gross and Microscopic Images:



Figure 1. Blood from oral cavity.

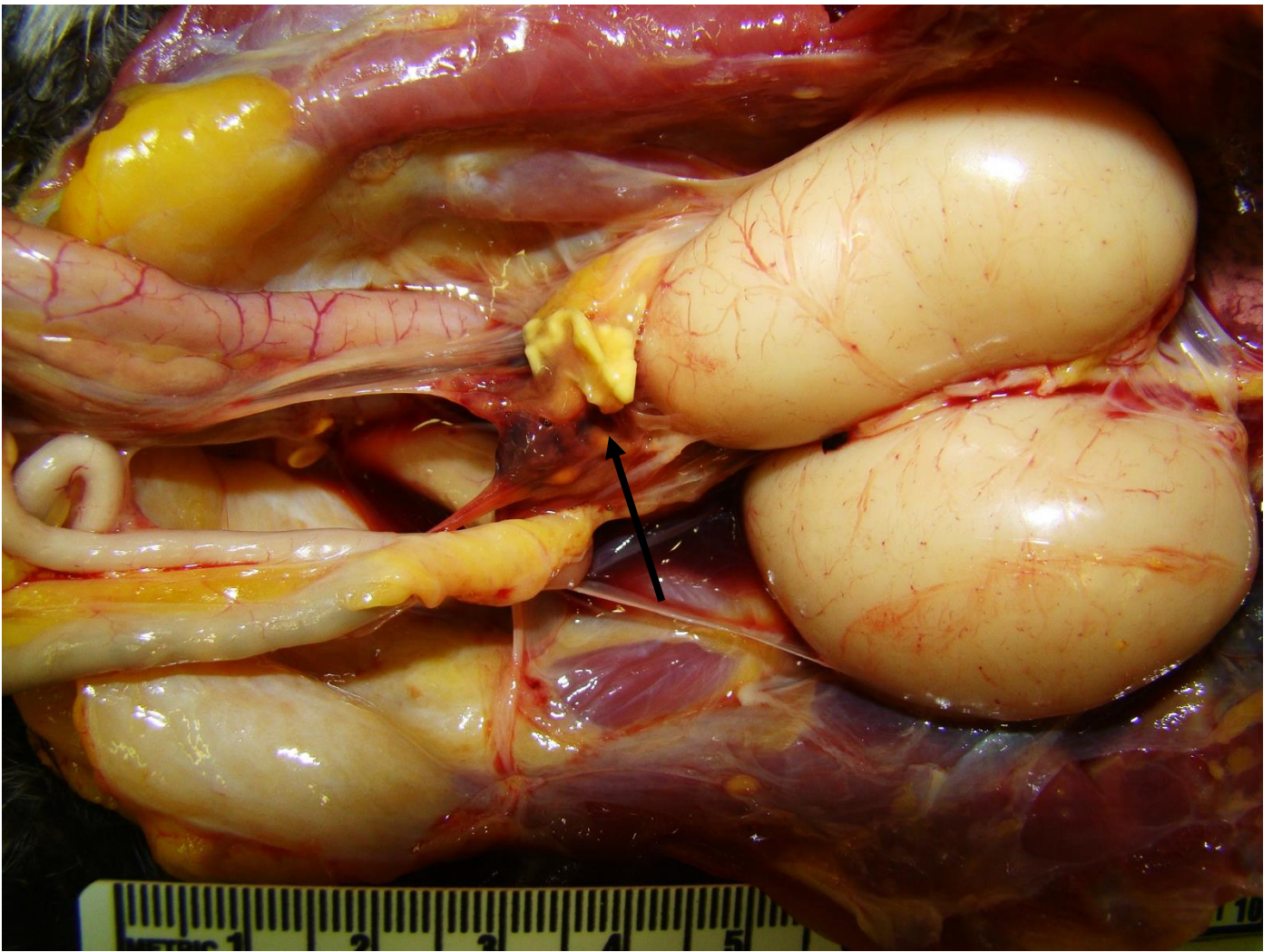


Figure 2. A granuloma (arrow) is adhered to the testicular blood supply.

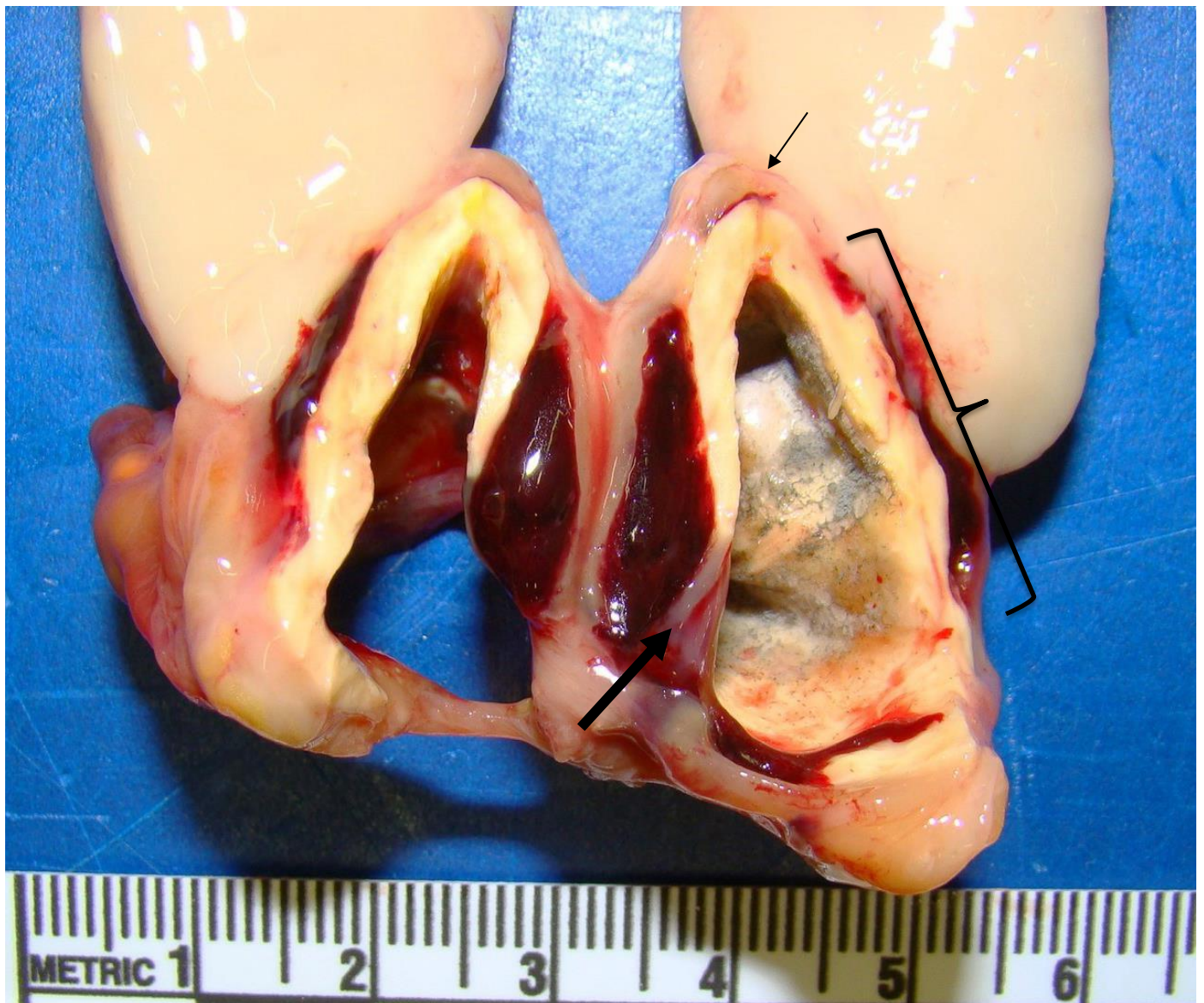
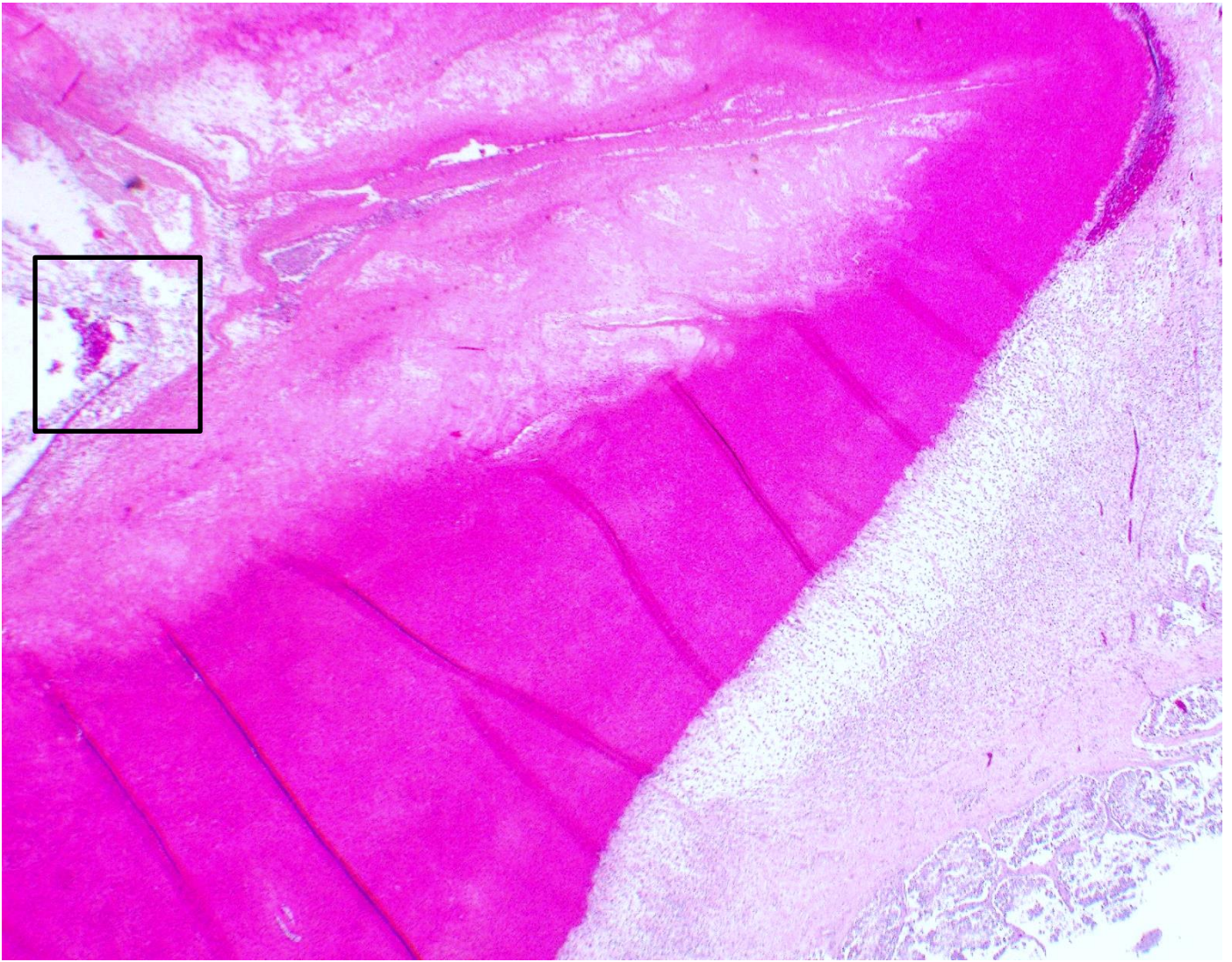
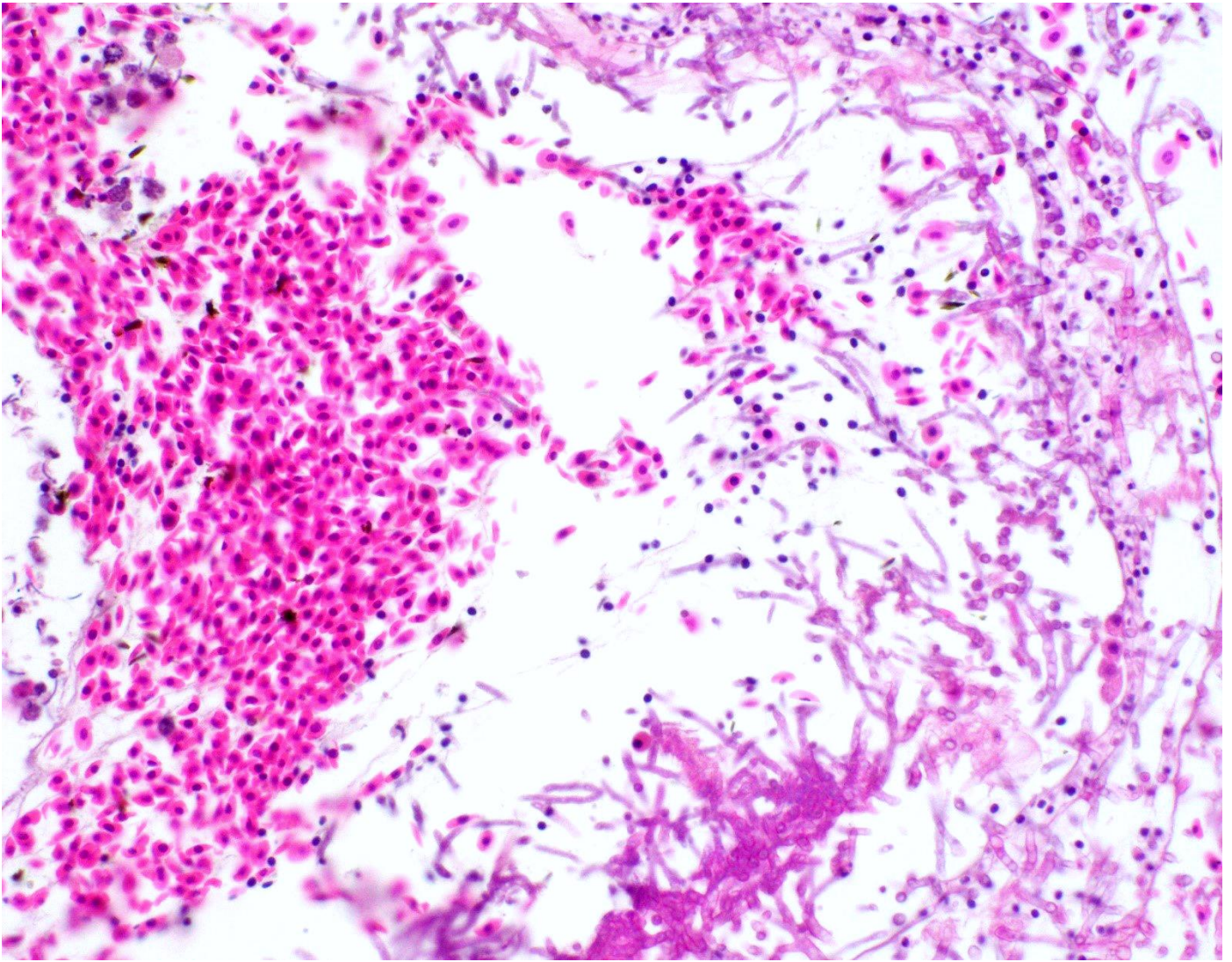


Figure 3. The granuloma (bracket) is adhered to the testicular tunic (thin arrow). It has a thick yellow fibrous wall that is lined by gray to white fungal hyphae with invasion into the wall of the adjacent blood vessel (thick arrow).





Figures 4 and 5 (inset): Large granuloma with outer fibrous band and thick eosinophilic layer of degenerate heterophils and necrotic debris (Figure 4), lined inside by mats of fungal hyphae, inflammatory cells, and nucleated red blood cells (Figure 5, inset). 20X and 400X, H&E.

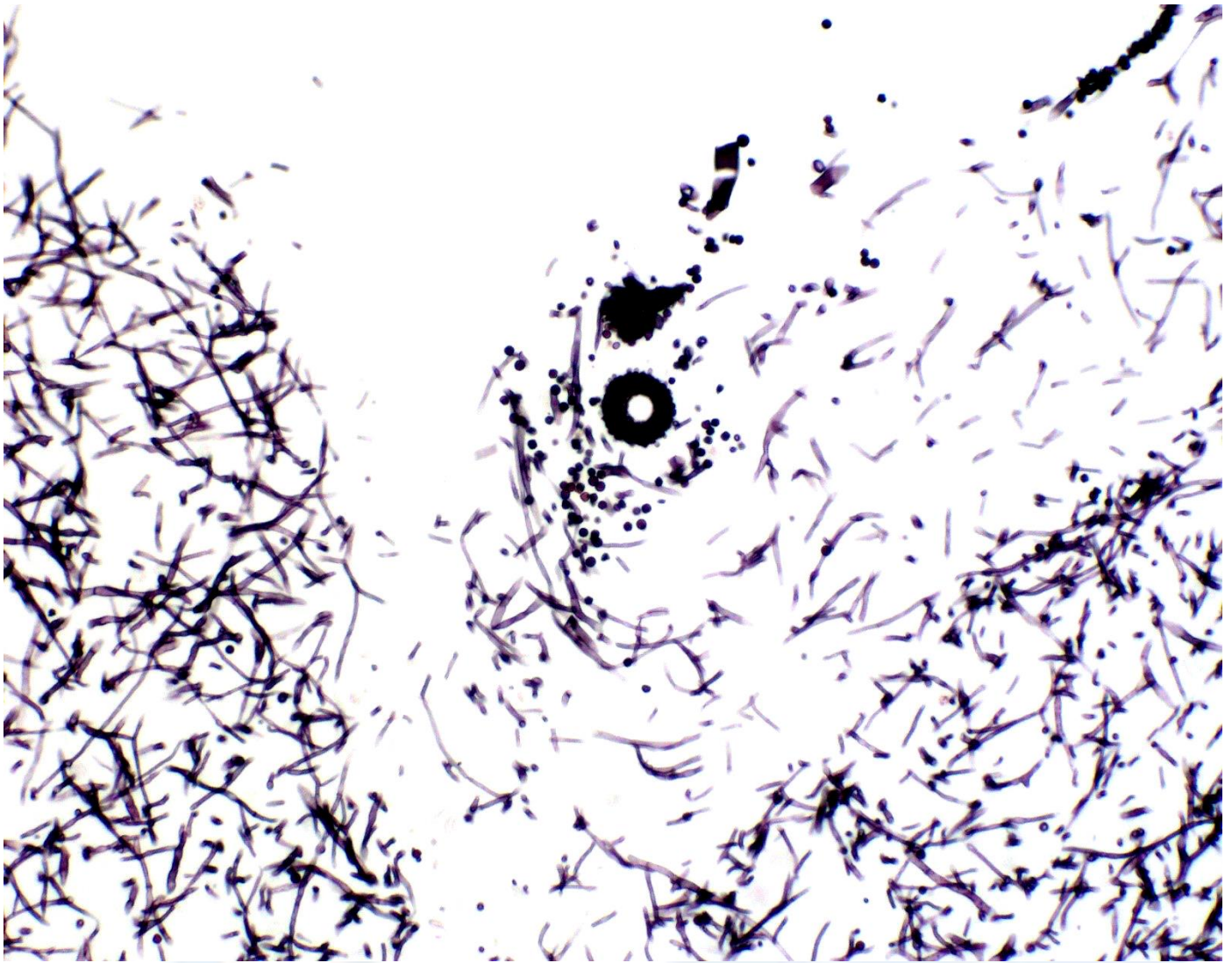


Figure 6: Fruiting bodies with spores and surrounding hyphae are consistent with *Aspergillus* sp. 400X, Gomori Methenamine-Silver (GMS).

Histologic Description: Testicular tunic: A 2 cm granuloma is adhered to the testicular tunic. The granuloma is composed of a thick fibrous capsule that has an outer layer of compact mature fibroblasts and an inner layer of hypertrophic (reactive) fibroblasts that are separated by increased clear space (edema), mixed with small caliber blood vessels and scattered transmigrating heterophils. The inner layer is brightly eosinophilic and composed of abundant degenerate and viable heterophils, nucleated erythrocytes, large varisized areas of coagulative necrosis, fibrin, edema, and numerous septate hyphae with parallel 3 to 5 μ m thick walls, acute angle dichotomous branching, and terminal bulbous swellings that stain black with GMS. Fungal hyphae are more easily viewed on the inner wall, often forming 30 μ m thick mats that are mixed with nucleated erythrocytes and scattered heterophils and other inflammatory cells that are rarely bi or multinucleated.

Morphologic Diagnosis:

1. Testicle, tunic: Periorchid granuloma, focal, with fungal hyphae consistent with *Aspergillus* sp.

Etiology:

Aspergillus fumigatus
Other *Aspergillus* spp.

Comments:

The genus *Aspergillus* is composed of more than 300 species, a fraction of which are involved in animal or human infections mostly following environmental exposure. Various risk factors for infections in veterinary medicine include unhygienic management, trauma, anatomical conformation of the skull, or suspected immunological deficiencies. In animals, aspergillosis is mostly sporadic, but it can involve an entire flock on poultry farms or hatcheries (Elad et al. 2018). Commonly known conditions in veterinary medicine include disseminated canine aspergillosis, canine and feline sino-nasal and sino-orbital aspergillosis, guttural pouch mycosis in horses, mycotic abortion in cattle, mycotic keratitis in horses, and avian aspergillosis (Elad et al. 2018). Heavy fungal loads may be the source of massive poultry infections, whereas trauma is usually assumed to cause ophthalmic infections. Disseminated and rhino-nasal/rhino-orbital aspergillosis seem to be associated with breed predisposition related to immune deficiencies and skull conformation, respectively.

Avian aspergillosis can affect wild or domesticated birds that are typically young. Penguins seem to be more susceptible than other birds. Acute cases, such as in young birds, are typically due to high environmental spores; in older birds, husbandry issues are at fault. Fungal spores must be able to grow at higher temperatures since the average body temperature of birds is 40°C.

Aspergillus fumigatus is the most common isolate, but *Aspergillus niger* has also been found in wild birds (Kiser et al. 2018). *Aspergillus niger* is associated with oxalosis, a pathologic process when some *Aspergillus* species produce oxalic acid, which reacts with blood or tissue calcium to precipitate calcium oxalate (Pabuççuoğlu 2005). The anatomy of the bird predisposes them to infection since hollow air sacs within the bone and other body compartments facilitate the spread. Spores that reach the lower respiratory tract can be disseminated throughout the body. Clinical signs typically show respiratory distress, lethargy, inappetence, diarrhea, wasting, and feather ruffling. Lesions are most commonly found in the lungs and air sacs (Elad et al. 2018). In this bird, the fungus may have invaded the testicular vasculature, leading to terminal intracoelomic accumulation of blood, which then dispersed through the air sacs into the oral cavity.

Several diagnostic techniques are available, if warranted based on the value of the bird. MRI, CT, radiology and endoscopy can be helpful in identifying lesions. Detection of fumigaclavin, a fungal toxin, is currently being researched. Immunohistochemistry in the form of polyclonal anti-*Aspergillus* rabbit antibodies shows promise as a diagnostic method (Elad et al. 2018). In one study, infected quails had higher plasma levels of ceruloplasmin, unsaturated iron-binding capacity (UIBC), iron, and total iron-binding capacity (TIBC), and lower levels of haptoglobin compared with uninfected controls (Goetting et al. 2013). Antifungal treatment may be impractical since granulomata are typically walled off from the blood supply. Nanoparticulate aerosol administration has shown promise in getting exposure while avoiding toxic doses but nebulized drug administration has yet to be well characterized in birds (Rundfeldt et al. 2013).

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