



Latin Comparative Pathology Group

The Latin Subdivision of the CL Davis Foundation

Diagnostic Exercise

Case #: 20 Month: April Year: 2012

Answer Sheet

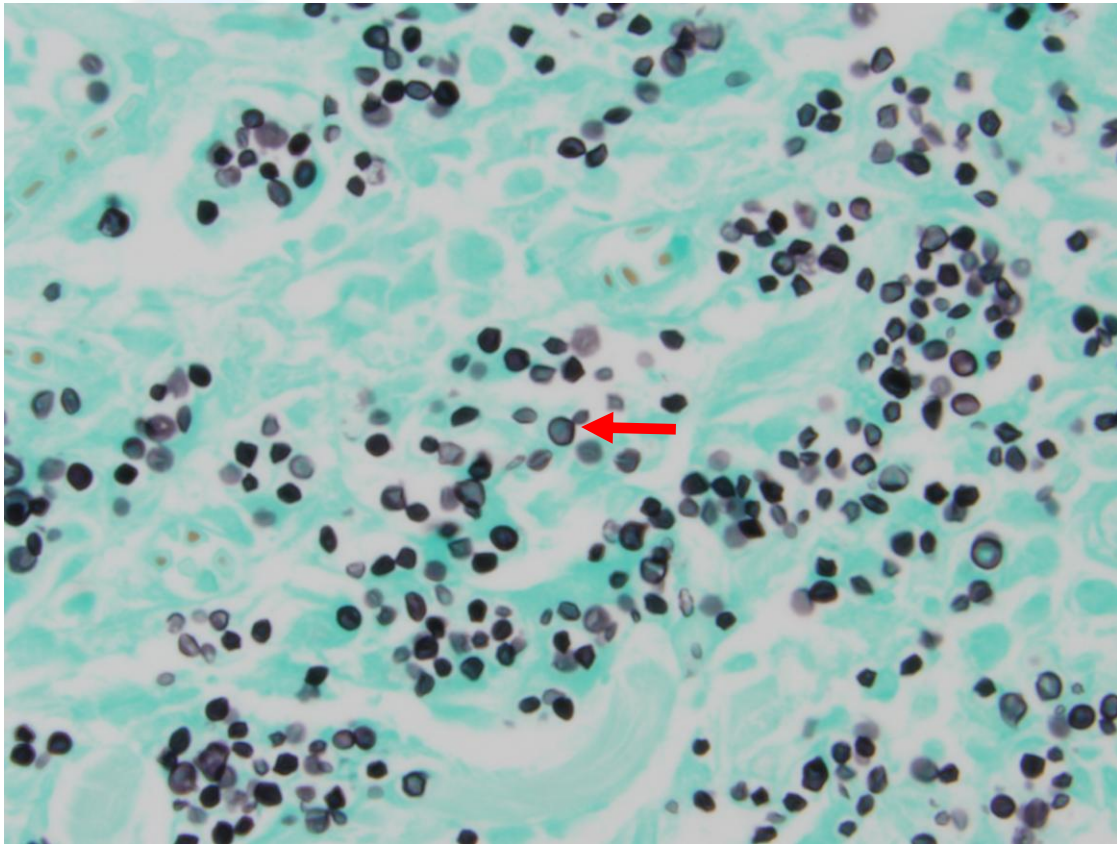
Gross findings:

On post mortem examination there were multiple exudative ulcers on the skin of the nasal planum, tail, left paw and external surface of the left pinna as well as on the dorsal surface of the tongue. The left mandibular lymph node was enlarged and presented purulent content when sectioned. Both nostrils were partially obstructed by a severe edema of the nasal mucosa, which was also ulcerated. There were no gross findings in the other organs that indicated systemic involvement.

Microscopic findings:

Microscopic examination revealed pyogranulomatous ulcerative dermatitis, panniculitis, localized myositis, rhinitis, glossitis and lymphadenitis of the left mandibular lymph node. In all these affected areas there were abundant yeasts, measuring 4–6 μm in diameter, surrounded by a clear halo and presenting eosinophilic cytoplasm. These yeasts were found mainly in the cytoplasm of macrophages, but were also extracellular in the purulent exudate and epidermal crust. There was also edema in the lungs. The Grocott's methenamine silver (GMS) stain (Figure 1) revealed abundant dark staining, 4–6 μm in diameter, cigar-shaped or oval yeasts in the dermis, subcutaneous tissue and subjacent skeletal muscles, nasal mucosa and tongue. In the lungs, rare yeasts were observed within alveolar macrophages. Some of the yeasts exhibited single budding with a narrow base (arrow), which is a characteristic of the fungus *Sporothrix*.

Figure 1. Grocott's methenamine silver stain, 60x



Laboratory testing:

The fungus *Sporothrix* was isolated through mycological culture of a swab from the ulcerated lesion on the external surface of the left pinna.

Diagnoses:

1. **Morphologic diagnosis:** Haired skin of nasal planum: Severe, chronic, ulcerative, diffuse pyogranulomatous dermatitis.
2. **Etiologic diagnosis:** *Sporothrix* dermatitis.
3. **Differential diagnosis:**

Histopathological findings observed in hematoxylin and eosin (H&E)-stained tissue sections are not disease-specific and can also be seen with other pathogenic fungi and protozoa. The main histologic differential diagnosis is histoplasmosis. Others are leishmaniasis, toxoplasmosis and cryptococcosis.

Discussion:

Sporotrichosis is a mycotic disease of worldwide occurrence caused by the thermal dimorphic fungus *Sporothrix schenckii*, now named *S. schenckii* species complex, which infects humans and many animal species. Recent molecular studies have shown that there are multiple species in the *Sporothrix schenckii* complex: *S. albicans*, *S. brasiliensis*, *S. globosa*, *S. luriei*, *S. mexicana* and *S. schenckii*. The species *S. brasiliensis* is considered the most virulent followed by *S. schenckii* and *S. globosa*.

In nature, the fungi of the *S. schenckii* species complex are saprophytes of senescent or dead vegetation and soil rich in decaying organic matter. Generally, the infection results from direct inoculation of the fungus into the skin via contact with contaminated plants or soil; less frequently, infection can also result from inhalation of conidia. Additionally, zoonotic transmission through bites or scratches of animals has been described in isolated cases or small outbreaks. However, an endemic form of sporotrichosis in which the domestic cats are the main source of infection for humans emerged in 1998 in Rio de Janeiro, Brazil. In this epidemic region, thousands of sporotrichosis cases in humans and cats and hundreds of such cases in dogs have been reported. The transmission from cats to other animals and to humans occurs through scratches, bites or contact with the exudate of their infected wounds.

Following inoculation, the fungus penetrates into deeper layers of tissue where it converts into yeast. It can remain in the dermis and subcutaneous tissue at the inoculation site, spread up through regional lymphatics and produce lymphangitis and lymphadenitis and disseminate systemically through blood vessels. In cats, the inhalatory route seems to be also important due to the high frequency of respiratory signs and pulmonary and nasal mucosal lesions associated with *Sporothrix*. Multiple skin lesions in cats can occur because of self-trauma, grooming and hematogenous dissemination from the lungs or perhaps from the initial skin lesion.

Cats are highly susceptible to sporotrichosis and systemic involvement is frequent in this species, leading to serious, difficult-to-treat forms of the disease and to death. This is different from what occurs in humans and dogs, in which the disease is essentially benign, although rare osteoarticular and disseminated forms can develop also in these species. In humans and dogs the response to treatment is usually good, while this is generally not the case in cats. The causes of the high susceptibility of cats to sporotrichosis are still unknown. The disease does not seem to be associated with infection with the feline immunodeficiency virus (FIV) or the feline leukemia virus (FeLV). Presumably because of the behavior of fighting for their territory, a higher prevalence of disease is found in adult male and sexually intact animals

The most common gross lesions in cats are multiple skin nodules, which can drain purulent or seropurulent content and exudative ulcers with frequent mucosal (conjunctival, nasal, oral, or genital) involvement. Pyogranulomatous lymphadenitis and extensive areas of necrosis in the skin that expose muscle and bone can also be observed. The most affected body regions are the head, especially the nose and ears, the tail and the hind limbs. Histologically, the cutaneous lesions of feline and canine sporotrichosis are characterized by ulcerative and variable but usually intense pyogranulomatous inflammatory reaction in the dermis, which can reach the panniculus and subjacent skeletal muscles. Yeasts can be seen inside macrophages and neutrophils and extracellularly in the purulent exudate and epidermal crust. There is an inverse correlation between granuloma formation and number of yeasts in dogs and cats. In cats, different from humans and dogs, the low frequency of granulomas and the great quantity of yeasts in the lesions reveal their higher susceptibility to sporotrichosis. Multinucleated giant cells are infrequent in dogs and cats compared to their occurrence in humans. Unlike dogs and humans, systemic involvement is common in cats with mild to moderate mixed inflammatory infiltrate of polymorphonuclear and mononuclear cells and yeasts observed histologically in lungs, liver, spleen, eye, kidney, adrenal glands and lymph nodes.

The definitive diagnosis of sporotrichosis depends on fungal isolation in culture media or Polymerase Chain Reaction (PCR). Nonetheless, cytology, histopathology, and immunohistochemistry are very useful routine tools for the preliminary diagnosis. In histopathology, specific histochemical staining techniques such as GMS and periodic acid Schiff (PAS) are needed to identify the yeasts of *Sporothrix*. The yeasts suggestive of *Sporothrix* are oval or cigar-shaped, range in size from four to six μm and generally exhibit single budding with a narrow base. Samples for laboratory diagnosis include swabs of the nasal cavities or exudative lesions, purulent or seropurulent content aspirated from nonulcerated abscesses, and incisional skin biopsy specimens.

Recommended literature:

- 1- Barros MBL, Almeida-Paes R, Schubach AO. *Sporothrix schenckii* and sporotrichosis. Clin Microbiol Rev. 2011; 24(4): 633-654.
- 2- López-Romero E, Reyes-Montes MR, Pérez-Torres IA et al. *Sporothrix schenckii* complex and sporotrichosis, an emerging health problem. Future Microbiol. 2011; 6(1): 85-102.
- 3- Miranda LHM, Quintella LP, Menezes RC et al. Evaluation of immunohistochemistry for the diagnosis of sporotrichosis in dogs. Vet J. 2011; 190 (3): 408-411.
- 4- Miranda LHM, Quintella LP, Santos IP et al. Histopathology of canine sporotrichosis: a study of 86 cases from Rio de Janeiro (2001-2007). Mycopathologia. 2009; 168: 79-87.

- 5- Miranda LHM, Quintela LP, Santos IB et al. Comparative histopathological study of sporotrichosis and american tegumentary leishmaniosis in dogs from Rio de Janeiro. *J Comp Path.* 2010; 143: 1-7.
- 6- Schubach TP, Menezes RC, Wanke B. Sporotrichosis. In: Greene CE. *Infectious Diseases of the Dog and Cat*, 4th ed. USA: Saunders, 2012. p. 645-650.
- 7- Schubach TMP, Schubach AO, Cuzzi-Maya et al. Pathology of sporotrichosis in 10 cats in Rio de Janeiro. *Vet Rec.* 2003; 152: 172-175.
- 8- Schubach TM, Schubach A, Okamoto T et al. Evaluation of an epidemic of sporotrichosis in cats: 347 cases (1998-2001). *J Am Vet Med Assoc.* 2004; 224(10): 1623-1629.

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A final document containing this material with answers and a brief discussion will be posted on the C. L. Davis website by the end of the current month (http://www.cldavis.org/lcpg_english.html).