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Cryptococcosis in a Persian cat: Cytological and cultural confirmation of *Cryptococcus neoformans*

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Abstract

Cryptococcus neoformans infection was identified in a five-year-old female Persian cat presenting with lesions on the nasal planum, based on cytological and cultural findings. Cytological evaluation of impression smears demonstrated characteristic budding yeasts with a soap-bubble appearance and mycological culture yielded smooth, mucoid, creamy-white colonies on Sabouraud's Dextrose Agar. The findings emphasize that feline cryptococcosis should be included in the differential diagnosis of cats with nasal or facial lesions even in the absence of respiratory signs, and that accurate cytological, cultural, and advanced diagnostic methods, along with restricted outdoor access, avoidance of avian-dropping contaminated environments and regular follow-up with continued antifungal therapy, are essential for optimal patient care, epidemiological understanding, prevention of recurrence, and sustained recovery in affected cats.

Keywords: Cat, feline, feline yeast infection, cryptococcosis, *Cryptococcus neoformans*, fungal culture, cytological findings

Introduction

Cryptococcosis is the most common systemic fungal disease of cats worldwide and is caused by members of the *Cryptococcus neoformans*-*Cryptococcus gattii* species complex, which comprises eight genotypes and several subtypes (strains) exhibiting distinct geographical distributions, pathogenicity, and antifungal susceptibility profiles (Pennisi *et al.*, 2013) [1]. The authors furthermore stated that the disease may manifest in nasal, central nervous system (either arising from nasal involvement or occurring independently), cutaneous or systemic forms.

From an ecological standpoint, avian guano, particularly pigeon droppings and decaying vegetation substrates such as *Eucalyptus* leaves are recognized risk factors for cryptococcal infection (Fortes *et al.*, 2001) [2]. Kwon-Chung *et al.* (2002) [3] also reported that pigeon droppings and soil contaminated with avian guano constitute the principal natural reservoirs of *Cryptococcus neoformans*. In this regard, Markey *et al.* (2013) [4] have stated that *Cryptococcus* organisms can persist in avian feces for up to one year, as the high creatinine content suppresses competing microorganisms and provides favorable conditions for the survival and proliferation of *Cryptococcus* spp.

The predominant clinical manifestations of feline cryptococcosis include sneezing, nasal discharge, epistaxis, respiratory distress, submandibular lymph node enlargement and cutaneous lesions such as erythema, erosions, ulceration and solitary to diffuse facial swelling, primarily around the nasal region. The major differential diagnosis of cutaneous form of cryptococcosis in cats include Feline Eosinophilic Granuloma complex (FEGC) which also exhibits a spectrum of dermatopathological lesions such as plaques, nodules, granulomas or ulceration in cats. FEGC is not a fatal disease or condition of cats but recurrence of the condition after treatment, complications due to secondary bacterial infection of the exposed skin lesions and delayed healing of the lesion due to frequent scratching of the lesion in an attempt to alleviate pruritus and continued presence of the source of allergy such as insects in the vicinity of cats are the reasons of worry while treating and managing a case of Feline

Eosinophilic Granuloma complex (Gowri *et al.* 2022) [5]. In this perspective, the authors suggested diagnosis of Feline Eosinophilic Granuloma complex in cats with dermatopathological manifestations by microscopic examination of direct impression smears and/or histopathology samples from the lesions and after ruling out other dermatopathological conditions in cats.

Compared with other pet and domestic animals, cryptococcosis occurs more frequently in cats due to their higher susceptibility to the pathogen, thereby allowing them to act as sentinels for monitoring potential human exposure to the fungus (Sykes and Malik, 2012) [6]. McGill *et al.* (2009) [7] reported that cats are five to six times more likely to be affected than dogs and approximately three times more likely than horses.

Cytological examination of samples, including cutaneous lesions, growths, lymph nodes, body fluids such as pleural fluid/Bronchoalveolar Lavage Fluid (BALF), cerebrospinal fluid, along with mycological culture is commonly employed for the diagnosis of feline cryptococcosis. Pennisi *et al.* (2013) [1] highlighted cytology as a convenient diagnostic tool, noting that the organisms have a characteristic appearance and are typically present in high numbers within lesions.

Histopathological examination of biopsy samples from suspected lesions and associated lymph nodes in cats can aid in identifying fungal structures and assessing histological changes in cryptococcosis. Additionally, immunohistochemistry and special stains such as mucicarmine, India ink, Periodic Acid-Schiff (PAS) and Grocott's methenamine silver, may be used in cases of special interest. Advanced diagnostic modalities for feline cryptococcosis include imaging techniques, MALDI-TOF mass spectrometry (Matrix-Assisted Laser Desorption/Ionization-Time-of-Flight) and the Cryptococcal Antigen Latex Agglutination System (CALAS) test.

With this background, this paper reports a case of cutaneous form of *Cryptococcus neoformans* infection in a cat, diagnosed by cytological and cultural investigations, and its post-therapeutic outcome.

Materials and methods

A five years old, female, Persian cat was presented with irregular, ulcerated lesions in the nasal region. In anamnesis, difficulty in breathing or nasal discharge was not reported. Clinical samples such as blood, serum, impression smears, fine needle aspirates, crusts, scabs etc. were collected for a detailed clinicopathological investigation.

Impression smears and fine needle aspirates obtained from multiple grossly affected areas of the nasal region were air-dried and stained using the Leishman-Giemsa (LG) technique. The stained preparations were then examined microscopically to identify specific etiologic agents, including bacteria, as well as fungal elements such as spores and hyphae.

Crusts and scabs collected from the nasal lesions of the affected cat were cultured on Sabouraud's Dextrose Agar (SDA) and Dermatophyte Test Medium (DTM) to facilitate the isolation and identification of potential fungal etiological agents. The plates were incubated at 25°C and 37°C for 4-6 weeks to permit fungal colony development. Culture plates were inspected on both the obverse and reverse sides on alternate days to document gross colony characteristics such as colour, texture, margin morphology and growth rate. For microscopic identification, selected colonies were collected and stained with lactophenol cotton blue (LPCB) and

examined under low and high-power magnification to identify characteristic fungal structures, including budding cells, sporangia and conidia (Subapriya *et al.*, 2015) [8].

Results and Discussion

Cytological evaluation of the suspected lesions showed round to oval basophilic budding yeast cells with a clear halo and a thin to barely discernible, indistinct capsule, imparting the typical soap-bubble appearance characteristic of *C. neoformans* (Figs. 1, 2). The fungal elements were observed against a background of numerous red blood cells.

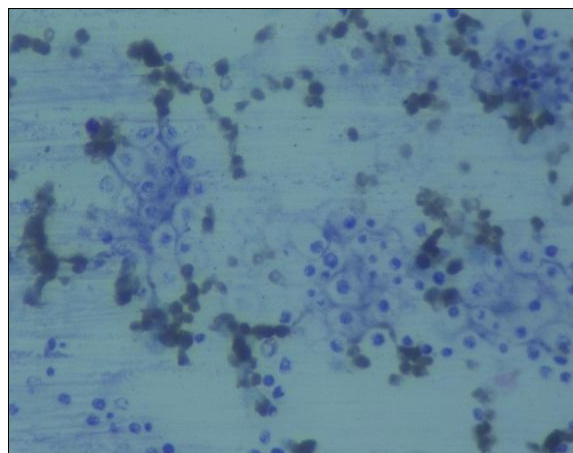


Fig 1: Cytology - *Cryptococcus neoformans* - Soap bubble appearance - RBCs - LG x 100

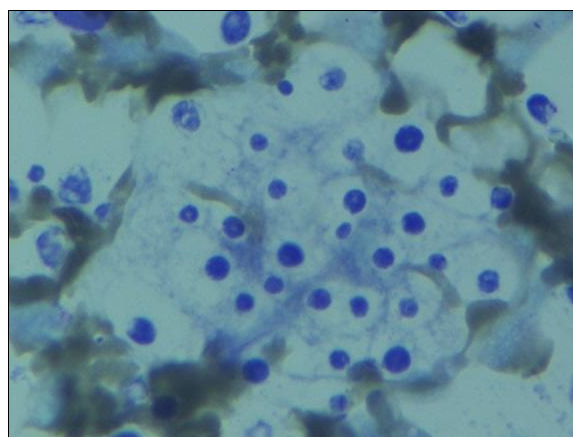


Fig 2: Cytology - *Cryptococcus neoformans* - Soap bubble appearance - RBCs - LG x 1000

Fungal culture on Sabouraud's Dextrose Agar (SDA) yielded white, creamy colonies of *C. neoformans* after three days of incubation. Microscopic examination of the colonies using Lactophenol Cotton Blue (LPCB) staining demonstrated round to oval budding yeast cells.

Based on the cytological findings, cultural characteristics, and microscopic examination of lactophenol cotton blue (LPCB)-stained smears, cryptococcosis was diagnosed in the cat presented for clinicopathological examination. Post diagnosis, the cat was treated with itraconazole following which the cat had a successful recovery. The pet parent was advised to present the cat for biweekly follow-up examinations over a period of two months to monitor for any recurrence of clinical signs.

Our study confirmed cryptococcosis in a five-year-old female Persian cat which is in partial corroboration with the findings of Trivedi *et al.* (2011) [9] who reported that although cats of any age may be affected, the median age of cats with

cryptococcosis is six years, with young adult cats appearing to be at increased risk.

Richards and Green (2006) [10] reported that cryptococcosis is the most common systemic fungal infection in cats and should be included in the differential diagnosis for cats presenting with respiratory tract disease, subcutaneous nodules, lymphadenopathy, intraocular inflammation, fever, or central nervous system involvement. In the present case, respiratory distress was not observed, and cytological examination of lymph node smears did not reveal any fungal etiological agents, suggesting a cutaneous form of feline cryptococcosis. Therefore, while vigilant evaluation for nasal cryptococcosis is warranted in cats presenting with respiratory signs, cats presenting with cutaneous lesions, particularly in the nasal region, should also be thoroughly investigated for cutaneous cryptococcosis, regardless of the presence or absence of respiratory signs, using appropriate clinicopathological methods.

Conflicting postulates exist regarding the zoonotic aspects of feline cryptococcosis. Irrespective of its zoonotic significance, accurate diagnosis of feline cryptococcosis based on cytological and mycological investigations, along with appropriate therapeutic intervention, is imperative for restoring the health and wellness of affected cats. Furthermore, confirmation of cryptococcosis through appropriate clinicopathological and advanced diagnostic techniques in suspected clinical cases will aid in generating epidemiological data to estimate the prevalence of the disease in the feline population and the environment. In concordance with this, Reis *et al.* (2021) [11] reported that animals may serve as sentinel hosts and that investigation of environmental sources prompted by feline cases is valuable for implementing interventions to reduce the risk of infection in both humans and animals.

Conclusion

Our study highlights the successful diagnosis and management of *Cryptococcus neoformans* infection in a Persian cat presenting with cutaneous lesions on the nasal planum. Cytological examination and fungal culture confirmed the presence of characteristic budding yeast cells, and treatment with itraconazole resulted in complete clinical recovery, emphasizing the importance of early diagnosis and appropriate antifungal therapy.

The findings further underscore that feline cryptococcosis should be considered in the differential diagnosis of cats with nasal or facial lesions, even in the absence of respiratory signs, and that accurate cytological, cultural, and advanced diagnostic methods are essential for optimal patient care and epidemiological understanding, reinforcing the role of cats as sentinel hosts for environmental exposure to *C. neoformans*. Restricting outdoor access of cats, particularly avoiding dusty environments contaminated with avian droppings, along with regular follow-up and continuation of antifungal therapy, is essential to prevent recurrence and ensure sustained recovery in affected cats.

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Conflict of interest

Not available

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Not available

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