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Original article

Fungal rhinitis in dogs

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Abstract

Fungal rhinitis and sinusitis in dogs are quite common reasons of chronic nasal discharge and rhinoscopy in such cases is commonly suggested. Forty three dogs were examined using rhinoscopy because of the presence of chronic airway symptoms. Clinical examination, routine hematology and serum biochemistry profiles, nasal and frontal sinus radiographs were made in all animals. Additionally, computed tomography in one dog was performed. Samples for histopathology were taken from 9 patients during rhinoscopy, additionally, from 8 of these patients samples for cytopathology were collected by blind nasal swab technique. In 9 of 43 dogs (20,5%), all males aged 1 to 13 years, examinations led to a diagnosis of fungal rhinitis. In 2 cases a diagnosis of fungal rhinitis was obtained based solely on cytopathology, while in 7 cases – mycosis of nasal mucosa was confirmed by histopathology. The present study revealed that cytopathological examination of nasal swabs has a low diagnostic value in the case of nasal infections in dogs. Although, in some dogs cytopathology, together with other widely available diagnostic techniques was sufficient to reliably diagnose fungal rhinitis, histopathology of samples collected during rhinoscopy is still the gold standard in such cases.

Key words: dog, fungal infection, nasal discharge, rhinitis, rhinoscopy

Introduction

Fungal rhinitis and sinusitis in dogs are quite common reasons for chronic nasal discharge (Sharp et al. 1991, Meler et al. 2008). Mesocephalic and dolichocephalic young or middle-aged dogs are most commonly affected. The most important cause is *Aspergillus fumigatus,* although *Aspergillus flavus, Aspergillus niger, Aspergillus nidulans, Penicillium spp, Cryptococcus neoformans, Alternaria* or *Rhinosporidium* could also cause the disease (Żmudzka and Lechowski 2008). Most important symptoms suggesting fungal rhinitis are chronic seropurulent or mucopurulent discharge, intermittent bouts of epistaxis, nasal pain, prolonged sneezing, ulceration and depigmentation of the nostrils (Peeters and Clercx 2008). Fungal infections of frontal sinuses and the nasal cavity cause severe damage of the mucosa and turbinates as a result of necrosis and/or proliferation secondary to vasculitis, toxic influence of fungal metabolites and inflammatory infiltrate. The most important differential diagnosis for fungal rhinitis is nasal neoplasia, so careful diagnostic evaluation should be performed in all cases (Johnson et al. 2006). Several diagnostic techniques can be used to confirm the diagnosis of fungal rhinitis, including radiography, computed tomography, magnetic resonance imaging, rhinoscopy and serology (Saunders and van Bree 2003,

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De Lorenzi et al. 2006, Johnson et al. 2006, Pomrantz et al. 2007, Billen et al. 2009). It should be underlined that fungal rhinitis is not always a primary disease and could be secondary to immunosuppression, foreign body presence, and rarely to neoplastic process. Samples for histopathological examination should be therefore taken, either under visual control or (infrequently) via blind biopsy (Willard and Radlinsky 1999, Sapierzyński and Żmudzka 2009). Definitive diagnosis of the nasal fungal rhinitis should be established on the basis of the histopathological examination of tissue samples collected during rhinoscopy.

The character of the pathologic process within the affected tissue, as well as the identification of the infection etiology, can be established by cytopathology. Microscopic examination of cytological samples collected from patients is cheap, easy to perform in most veterinary clinics and usually only slightly invasive. Moreover, results can be obtained after a short time period. Treatment of canine nasal fungal rhinitis is difficult, highly specific, can be expensive, commonly requires invasive methods of treatment (nasal cavity flushing, nasal cavity trepanation) and prognosis is not always favorable. Therefore, in such cases the main requirement for introduction of correct treatment is a precise and definitive diagnosis. We suggest that cytological examination made during rhinoscopy enable a rapid and accurate diagnosis and facilitate a specific, less or more invasive, antifungal therapy even during the same anesthetic episode. The aim of the study was to present clinical, radiographic and endoscopic data on canine fungal rhinitis and to examine the usefulness of nasal swab cytology as a tool for rapid diagnosis of these infections in dogs.

Materials and Methods

Forty three dogs were admitted to The Small Animal Clinic between 2009-2010, for rhinoscopic examination because of clinical signs suggesting chronic upper respiratory tract disease. All the patients were clinically examined. Routine hematology, serum biochemistry profiles, and nasal and frontal sinus radiographs under general anesthesia (GE type Prestige II) were performed. In one dog computed tomography was additionally performed (GE tomograph type Hi Speed CT/e plus). For radiographic and tomographic examinations, patients were positioned in sternal recumbency. Anterograde rhinoscopy with a rigid endoscope (XP 700/33 Nopa C) and nasopharyngoscopy with a flexible endoscope (XP 20 Olympus with 100 cm working length and 8 mm diameter) were performed during the same anesthesia. During rhinoscopy the following findings were recorded and considered to be suggestive for rhinitis: general appearance of the nasal mucosa (color, surface, hyperemia, fragility – bleeding after contact with rhinoscope), presence and character of exudates. Additionally, architecture of the nasal turbinates was assessed and the presence of deformity and/or ulceration and/or other abnormalities of mucosa and turbinates was recorded. Samples for cytopathology and histopathology were collected during rhinoscopy. Samples for cytopathology were collected directly after preliminary examination of affected nasal cavity/cavities. A sterile cotton swab was introduced into the nasal cavity, and then the material was transferred onto a microscopic slide by the rolling technique. Smears were then dried, fixed in 70% methanol, stained with Giemsa solution, and examined under light microscope. Changes were classified according to the presence of inflammatory cells (inflammation or non-inflammatory state), types and number of inflammatory cells (type of inflammation), the presence or absence of etiologic agents (aseptic, bacterial, fungal), and the evidence of necrosis and proliferation of epithelial cells. For histopathology samples of nasal mucosa, turbinates or pathologic masses were collected from the same dogs using flexible gastroscope forceps (Olympus XP20). In one dog material for histopathological examination was collected by the owner after intensive sneezing (turbinate fragment; in this case cytology was not performed). After collection, tissue samples were fixed in a 4% buffered formalin, and then processed using routine methods. Slides were stained with hematoxylin and eosin method and the PAS method (for identification of fungal hyphae). Based on the morphological criteria, a final histopathological diagnosis was made. Finally, results of cytopathology and histopathology were compared.

Statistical analysis. Sensitivity of cytology in diagnosing fungal rhinitis was estimated according to Thrusfield (2005). Histopathology was considered a gold standard. 95% confidence intervals for proportions were calculated using Wilson score method (Altman et al. 2000).

Results

Among 43 dogs examined by rhinoscopy during the study period, fungal rhinitis was recognized in nine animals – 20.9% of all patients (95% CI: 11.4%, 35.2%). Data on patients characteristics are presented in Table 1. Clinical signs suggestive of nasal disease were persisting from 2 weeks to 7 months. Direct indications for rhinoscopy included chronic mucopurulent/purulent nasal discharge or epistaxis, however

Table 1. Signalment, duration and character of clinical signs in nine dogs with fungal rhinitis.

No.	Patient	Nasal discharge	Epistaxis	Sneezing	Deformity or ulceration of nose/nasal planum
1.	German Shepherd, 4 years, ♂	Mucopurulent lasting 7 months	Couple Times	Intensive	Deformity of the right nostril
2.	Miniature Schnauzer, 4 years, ♂	Purulent, bilateral lasting 1.5 months	None	Mild	None
3.	Mixed, 7 years, ♂	Purulent lasting 2 months	None	Mild	Deformity of the nose bridge
4.	Golden Retriever, 11,5 years, ♂	Mucopurulent lasting 4 months	Twice	Mild	None
5.	Mixed, 13 years, ♂	Purulent lasting 2 months	None	Intensive	Ulceration of the nostrils area
6.	Golden Retriever, 1 year, ♂	Purulent lasting 6 months	None	Intensive	None
7.	Amstaff, 10.5 years, ♂	None	Couple times, severe	Mild	None
8.	Mixed, 6 years, ♂	None	Couple times	Mild	Deformity of the nose bridge
9.	Golden Retriever, 5 years, ♂	Mucopurulent lasting 5 months	Twice, severe	Intensive	None

Table 2. Cytopathological preliminary diagnoses and final histopathological diagnoses in nine dogs with fungal rhinitis.

No.	Cytopathological diagnosis	Histopathological diagnosis
1.	Purulent fungal rhinitis with marked epithelial cell atypia	Purulent and fungal rhinitis with marked epithelial cell atypia
2.	Purulent rhinitis and mild epithelial cell atypia	Purulent rhinitis with necrosis and fungal hyphae
3.	Epithelial cells with moderate atypia – suspicion of adenocarcinoma	Fungal rhinitis with purulent inflammation
4.	Purulent rhinitis; fungal hyphae with PAS method staining	Fungal rhinitis with purulent inflammation
5.	Bacterial purulent rhinitis mild epithelial cell atypia	Fungal rhinitis with purulent inflammation
6.	Rhinitis with mixed inflammatory infiltrate and necrosis – suspicion of fungal infection	Fungal rhinitis with mixed inflammatory infiltrate
7.	Rhinitis with mixed inflammatory infiltrate and necrosis – suspicion of fungal infection	Fungal rhinitis with mixed inflammatory and focally lymphoplasmacytic infiltrate
8.	Bacterial rhinitis with mixed inflammatory infiltrate and necrosis	Fungal rhinitis with mixed inflammatory infiltrate
9.	Not examined	Fungal rhinitis with mixed inflammatory infiltrate

sneezing of various intensity was also observed in every patient. In some dogs nose pain or discomfort of the facial region, stridor, ulceration of nasal planum or deformity of the nose bridge were recorded. Detailed data on symptoms and signs considered to be typical for fungal rhinitis are presented in Table 1.

Radiographic examination revealed increased tissue density of nasal cavities in every case (bilateral in 4 cases, unilateral in 5 cases), increased density of frontal sinus/sinuses in 6 cases (bilateral in 3 cases, unilateral in 3 cases), and nasal septum deterioration in one dog. Computed tomography performed in one patient showed more detailed findings, including turbinates atrophy, presence of mass with air caverns, nasal septum flexure and mucous membrane thickening. The rhinoscopic examination showed typical features of inflammation, including hyperemia, swelling and fragility of mucosa in all patients. Additionally, mucopurulent or purulent exudate covering the mucosal membranes as well as turbinates damage or atrophy were also recognized in all dogs. In 5 cases nasal fungal infection was suspected during rhinoscopy because of the presence of fungal plaques on the mucosal surface. Cytopathology of nasal swabs was

performed in eight of the nine patients. Based on the microscopic examination of the cellular samples cytological diagnosis of nasal fungal infection was established in 2 cases (based on the presence of fungal hyphae invading underlying tissues), in another 2 cases nasal mycosis was suspected, although microorganisms were not seen in microscopic slides. Therefore, sensitivity of cytology in diagnosing fungal nasal infection turned out to be low – only 25% (95% CI: 7.2%, 59.1%). Neutrophilic inflammation or mixed inflammatory infiltrates with features of tissue necrosis (abundant cellular detritus and proteinaceous material) were additionally present in these four cases. In the remaining four cases the presence of neutrophilic inflammation with numerous bacteria was observed and purulent bacterial rhinitis was the presumptive diagnosis. Additionally, mild to marked cellular atypia of epithelial cells were noted in half of the cases; cellular atypia was so marked in two cases that nasal adenocarcinoma/carcinoma was suspected. In all dogs histopathological examination revealed presence of fungal hyphae on the mucosal surface with severe purulent or mixed inflammatory infiltrates, commonly with tissue necrosis. Results of cytopathological and histopathological examinations in particular dogs are summarized in Table 2.

Discussion

The typical canine patient with a nasal fungal infection is a dolichocephalic or mesocephalic (commonly Golden Retriever, German Shepherd, Border Colli) breed, and a young (about 1 year old) or mature (more than 8 years old) dog. In most papers describing nasal fungal infections in dogs, similarly as in the presented study, the patients ranged from 1 to 13 years of age (Sharp at al. 1991). However, the median age of our patients was 6.8 years, which is slightly higher than in other studies (from 3.3 to 5 years of age) (Mathews 2004, Johnson et al. 2006, De Lorenzi et al. 2006, Meler et al. 2008). States of immunodeficiency or other factors including chronic antibiotic therapy, emaciation, or neoplastic disease can predispose the nasal mucosa to colonization and invasion by fungal hyphae. Impairment of the immune system in juvenile and elderly animals may predispose dogs to nasal mycosis, however, it is unclear whether impaired immune system (diminished function of lymphocytes) is the cause or the result of infection (Mathews 2004). Male dogs appear to be at greater risk than female dogs (Saunders at al. 2004, De Lorenzi et al. 2006) and this tendency was also seen in the present study – all the patients in this study were male. There is no simple explanation for this predisposition, possibly that nasal diseases are more prevalent in male dogs because of hormonal status or males are predisposed because of behavior (inquisitive) causing them to aspirate fungal spores during sniffing. Many of the dogs in this study were Golden Retrievers, in our opinion the cause of this was popularity of this canine breed, however breed predisposition to fungal rhinitis cannot be excluded.

According to literature the most typical clinical signs of fungal rhinitis in dogs include: purulent or mucopurulent nasal discharge, sneezing, nose pain or discomfort of the facial region, epistaxis, stridor, ulceration or depigmentation of external nares (Mathews 2004, Johnson et al. 2006, Lorenzi et al. 2006, Meler et al. 2008). In the present study, however, only sneezing was a consistent clinical manifestation. Chronic nasal discharge or/and epistaxis were recorded in a majority, but not in all patients. As our study has revealed, as few as two episodes of nose bleeding were enough to warrant rhinoscopy. In contrast, nasal discharge lasted from 1.5 to 7 months before rhinoscopy was performed and thus a diagnosis was reached late. It seems that epistaxis is a more alarming clinical finding, because it usually is considered to be related to nasal cancers or to raise suspicion of serious, potentially life threatening problems. Interestingly, maceration and depigmentation of the nasal planum, were not observed in dogs in the present study, although this clinical abnormality is thought to be typical for canine fungal rhinitis (Mathews 2004).

All the dogs in the present study were examined radiographically and one dog was examined using computed tomography. We found CT a much more useful tool than the classic X-ray in identification of pathological abnormalities, which is consistent with literature (Saunders at al. 2004). In all the presented cases, radiography showed only increased tissue density in the nasal cavity or paranasal sinuses, while only one dog showed additionally destruction of the nasal septum. Atrophy of turbinates, loss of trabecular pattern or other abnormalities considered to be typical for fungal rhinitis (Mathews 2004, Saunders et al. 2004, Meler et al. 2008) were not clearly visible during radiography in the present study. Tomography gives more information about turbinates and septum architecture and allows detection of atrophy, hypertrophy or deformation of turbinates (Mathews 2004, Saunders et al. 2004). CT distinguishes between the causes of increased density of the nasal cavity – the presence of fluid or mucosal thickening. Unfortunately, because of high costs of this visualization technique, it was only performed in one dog during this study. However, because it provides more detailed information, CT should be introduced in every possible case (Mathews 2004, Saunders et al. 2004).

For long time fungal cultures and serology were considered less useful for diagnosis of canine fungal rhinitis, because of a high number of false-negative results and the fact that some fungi (e.i. *Aspergilus flavus*) may be found as endogenous flora in the nasal cavity in many dogs (Sharp et al. 1991, Mathews 2004, Peeters and Clercx 2007, Żmudzka and Lechowski 2008). A recent study (Billen et al. 2009) suggests better sensitivity of fungal culture when samples are taken under visual control (endoscopy) and incubated in temperatures of 37 centigrades. However, results of mycology are available after a few days and may be ambiguous. Therefore, and in line with published data, we decided that rhinoscopy with histopathological examination of samples taken under eye control is still the best method to diagnose fungal rhinitis (Mathews 2004). Microscopic examination of tissues collected during endoscopy allow not only the detection of fungal hyphae, but also allow pathologist to decide if fungi invade superficial layers of nasal mucosa and if it is a primary disease or a process secondary to foreign body or neoplasia.

The diagnostic method that best ensures confirmation of fungal infection in dogs with the chronic nasal discharge is endoscopy with histopathology (Willard at al. 1999, Żmudzka and Lechowski 2008). The most important endoscopic sign suggesting fungal rhinitis is presence of a white coating covering the mucosal membrane. However, we have found such finding in only 5 of the 9 dogs examined by rhinoscopy. In the remaining cases this abnormality was not observed despite the presence of fungal hyphae invading the nasal mucosa during microscopic examination. It should also be mentioned that in dogs with nasal fungal disease, fungal plaques can be absent within the nasal cavity but can be present only in the frontal sinuses (Johnson et al. 2006). Although turbinated damage and necrosis of mucosa were found in all endoscopically examined dogs, these kinds of lesions are less diagnostically significant because they can be seen in other nasal diseases, e.g. neoplasms and severe cases of non-specific rhinitis.

The exudate covering the nasal mucosa or discharge commonly observed in dogs with chronic diseases of upper respiratory tract is a potential material for cytopathological examination. According to Mathews (2004) low-invasive modalities of specific antifungal therapy can be performed during the same anaesthetic episode after definitive, rapid diagnosis obtained on the basis of clinical history and radiographic findings supported by cytopathology. However, according to own observations and results of published studies, nasal discharge contains mainly abundant neutrophils, mucous and necrotic debris, thus such material is usually non-diagnostic (Mathews 2004, de Lorenzi et al. 2006). One of the purposes of the present study was to estimate usefulness of blind nasal swabs as a simple procedure of sample collection for identification of fungal nasal infection in dogs. In a study published by De Lorenzi et al. (2006) various nasal sample collection techniques for cytopathology were compared: blind swab collection, direct smears from the nasal discharge, brushing from suspected lesion under direct endoscopic visualization and squash technique of mucosal biopsies from suspected lesions obtained under direct endoscopic visualization. The most effective method of sample collection that contain fungal hyphae on microscopic slides were a squash technique (100% of cases) and brushing from suspected lesions (93,3% of cases examined), while the other two methods were less useful (direct smears of nasal discharge – 13.3% of cases; and blind swab collection – 20% of cases). However, procedures that are most useful can only be performed during endoscopic examination of the nasal cavities, unfortunately this diagnostic method is currently not widely available for veterinary patients, and can be expensive for some owners. On the other hand, a blind nasal swab collection procedure is possible in every veterinary clinic without specific equipment, and only mild sedation may be necessary in some patients. As the present study has revealed, cytopathological examination of nasal swabs collected was characterized by a low usefulness in diagnosis of nasal fungal infection in dogs. The presence of fungal hyphae within smears were rarely identified despite a quite abundant mass of fungal microorganisms observed during histopathological examination.

In some dogs cytopathology, together with other widely available diagnostic techniques, such as medical history and radiography, were sufficient to make a reliable diagnosis of fungal rhinitis. However, histopathologic examination of samples collected during rhinoscopy is still considered the gold standard in such cases.

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