# Multimodal treatment of recurrent sinonasal cryptococcal granulomas in a horse

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**Case Description**—A 7-year-old 509-kg (1,120-lb) Tennessee Walking Horse mare was evaluated because of bilateral mucosanguinous nasal discharge, intermittent right-sided epistaxis, and worsening dyspnea of 9 months' duration.

**Clinical Findings**—Multiple masses in the nasopharynx were detected via endoscopic and radiographic examinations. Cytologic and histologic examinations of biopsy specimens of 1 mass revealed round yeasts with thick nonstaining capsules and occasional narrow-based budding that resembled cryptococcal organisms.

**Treatment and Outcome**—Oral administration of fluconazole and organic ethylenediamine dihydriodide and intermittent intralesional injections with fluconazole, amphotericin B, and formalin resulted in resolution of lesions for a period of 2.5 years. The horse then developed exophthalmos, recurring clinical signs, and extensive nasopharyngeal masses. The masses were surgically debulked via a large frontonasal bone flap, and the horse was treated with IV injections of amphotericin B and long-term oral administration of fluconazole. Clinical signs did not recur in the following 2-year period. A presumptive diagnosis of cryptococcosis was made following cytologic and histologic evaluations of the masses; results of serologic analysis and fungal culture confirmed infection with *Cryptococcus neoformans*.

**Clinical Relevance**—Cryptococcal infection of the upper respiratory tract in horses has previously been described as a uniformly fatal disease. As this case report illustrates, medical and surgical treatment of sinonasal cryptococcal granulomas in horses may be successful, but the importance of long-term follow-up and the potential for disease recrudescence should be considered. As efficacious antifungal agents become less expensive, their increased use will likely decrease mortality rates in horses with fungal infections. (*J Am Vet Med Assoc* 2009;235:723–730)

In September 2003, a 7-year-old 509-kg (1,120-lb) Tennessee Walking Horse mare was examined at the John T. Vaughan Large Animal Teaching Hospital at the College of Veterinary Medicine, Auburn University, because of bilateral mucosanguinous nasal discharge, intermittent right-sided epistaxis, and progressive dyspnea of 9 months' duration. Periodic treatments with orally administered potentiated sulfonamides temporarily ameliorated clinical signs. Endoscopic examination of the right nasal passage was performed by the referring veterinarian; although epistaxis prevented completion of the examination, a mass was detected, which prompted referral of the horse to the teaching hospital.

At the initial evaluation at the hospital, the horse's heart rate, respiratory rate, and rectal temperature were within reference limits. Thoracic auscultation revealed stertorous upper airway sounds, although lung sounds were considered normal. Dried mucopurulent discharge was present around the right nostril, from which there was reduced airflow. Results of hematologic and serum biochemical analyses indicated mild anemia (PCV, 28.8%; reference interval, 32% to 48%) and neutrophilia (6.9 × $10^9$ cells/L; reference interval, 3 to 6 × $10^9$ cells/L).

Endoscopic examination of the nasal passages and skull radiography were performed. Endoscopy of both nasal passages revealed multiple yellow, mucus-covered masses that were located approximately 25 cm from the nares and that extended into the nasopharynx. In the right nasal passage, a much larger and darker mass was visible caudal to the smaller rostral masses. This mass obscured the right ethmoid turbinates and had the appearance of a progressive ethmoidal hematoma. With endoscopic guidance and by use of a mare uterine biopsy instrument,[a] a biopsy specimen was collected from a mass in the left nasal passage; the mass was located dorsal to the opening of the auditory tube diverticulum (guttural pouch; **Figure 1**). Because of the large size of the ethmoidal mass and the presence of multiple smaller masses rostrally, insertion of both the uterine biopsy instrument and the flexible endoscope in the right nasal passage was impossible; therefore, a specimen of the

large right-sided mass that had the appearance of an ethmoidal hematoma could not be obtained.

Cytologic and histologic examinations of the biopsy specimen revealed similar findings. Histologically, the specimen contained fibrovascular tissue that was infiltrated by lymphocytes, plasma cells, and numerous ovoid to elliptical (8 to 16 μm in diameter), lightly basophilic yeast cells surrounded by thick (8 to 12 μm) nonstaining capsules. In impression smears stained with modified Wright stain, the yeast had narrow-based budding typical of cryptococcal organisms[1] (**Figure 2**).

Skull radiography revealed a well-circumscribed, rounded soft tissue opacity over the ethmoid bone at the level of the right upper third molar tooth that was



Figure 1—Endoscopic view of a mass in the left nasal passage (25 cm from the nares) in a 7-year-old horse that was evaluated because of bilateral mucosanguinous nasal discharge, intermittent right-sided epistaxis, and worsening dyspnea of 9 months' duration. The mass was detected during an initial examination in 2003, as were several other masses in both nasal passages.
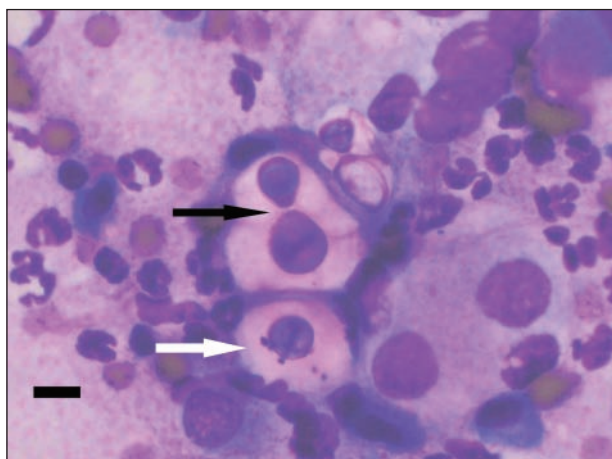


Figure 2—Photomicrograph of an impression smear prepared from a biopsy specimen of the mass in the left nasal passage of the horse in Figure 1. Cryptococcal organisms that are budding (black arrow) and that are surrounded by a clear unstained capsule (white arrow) are present. Modified Wright stain; bar = 10 μm.

most consistent with a progressive ethmoidal hematoma, although fungal granuloma was considered to be another potential cause of this lesion. The smaller masses in the nasal passages could not be identified on the radiographic views.

Thoracic radiography and a transtracheal wash were performed to rule out pulmonary mycotic infection. No abnormalities were detected via thoracic radiography. Cytologic examination of a tracheal wash fluid sample revealed mucus with entrapped neutrophils, many macrophages, and erythrocytes; however, no infectious agents were identified. Bacterial and fungal cultures of transtracheal wash fluid yielded no clinically relevant growth. Antifungal treatment with fluconazole[b] (14 mg/kg [6.4 mg/lb] loading dose, PO; followed by 5 mg/kg [2.3 mg/lb], PO, q 24 h for 6 weeks) was begun.[2]

The horse was reexamined 3, 10, and 15 weeks after the initial evaluation. At each of those recheck visits, intralesional treatments were performed during endoscopy; the horse remained in the hospital for 3 days after the first intralesional treatments. At weeks 3 and 10, masses with an appearance similar to the confirmed mycotic granuloma in the left nasal passage were injected with fluconazole[c] (total dose, 100 mg) by use of an endoscopic delivery catheter[d] (length, 190 cm; width, 2.5 mm) that was passed through the biopsy channel of a 1.5-m flexible endoscope. The large mass on the right side (tentatively diagnosed as a progressive ethmoidal hematoma) was injected with 60 mL of 10% formalin at weeks 3 and 10. Three days after the first injection of formalin, this mass had regressed in size considerably, and the surface was discolored and liquefied. It was injected with 10 mL of formalin at this time and again at week 10. Because of the response of the right-sided mass to the treatment with formalin, some of the left-sided masses were also injected with 10% formalin at this time and again on weeks 10 and 15. Organic ethylenediamine dihydriodide[e] (1.3 mg of active ingredient/kg [0.6 mg/lb] or 30 mg of the 4.57% dextrose powdered formulation/kg [13.6 mg/lb], PO, q 24 h for 4 weeks[3]) was administered in the horse's feed as an adjunctive antifungal treatment from week 3 to week 7.

By week 15, the nasal discharge had ceased and the horse had returned to its normal level of exercise according to the owners. At this time, all nasopharyngeal masses were reduced dramatically in size except for 1 mass within the right nasal passage (approx 8 cm rostral to the ethmoid turbinates) that was approximately 1 cm in diameter and that appeared slightly enlarged, compared with its size at week 10. Microscopic examination of an impression smear of this mass revealed numerous necrotic cells, neutrophils, and scattered columnar epithelial cells; mixed types of intra- and extracellular bacteria were observed, but no fungal organisms were identified. The bacteria were considered surface contaminants from the nasopharynx, and antibacterial treatment was not considered necessary. This mass was injected with amphotericin B[f] (50 mg) during the recheck visit, and all remaining smaller masses (0.5 cm in diameter) were injected with formalin.

In February 2005, the horse had been used successfully for pleasure riding for 1 year. Reexamination was performed to ensure the lesions had resolved completely or at least had not regrown substantially. During endoscopic examination of the nasopharynx, some scarring of the right ethmoidal region was seen. Cytologic

examination of an aspirate collected from a thickened area of mucosa cranial to the right ethmoid region revealed no abnormalities. In February 2006 (30 months after initial diagnosis), according to the owners, the horse was healthy and being used as a trail horse.

In May 2006, the horse was examined on the farm by the referring veterinarian, who was there to perform routine vaccinations. At that time, the owners stated that the horse had increasingly stertorous breathing, mucopurulent nasal discharge, and 2 episodes of right-sided epistaxis during the preceding 3-week period. A swab sample was collected from the right nasal passage for microbial culture and antimicrobial susceptibility testing. Cultures yielded *Staphylococcus intermedius* and 3 fungal organisms (*Cladosporium* spp, *Rhodotorula* spp, and *Cryptococcus* spp). The horse was again referred to the teaching hospital.

At the evaluation after admission to the hospital, the horse had a body condition score of 6 (scale of 9) and weighed 532 kg (1,170 lb); heart rate, respiratory rate, and rectal temperature were within reference limits. Exophthalmos of the right eye was obvious, although the horse appeared to have normal vision and no evidence of cranial nerve dysfunction. Ultrasonographic imaging of the right globe and orbit failed to identify any abnormalities. A CBC, fibrinogen concentration assay, and serum biochemical analyses were performed; the only abnormal finding was hyperglobulinemia (6.4 g/dL; reference interval, 2.8 to 4.4 g/dL). Skull radiography revealed soft tissue opacities in both the right and left nasal passages and in the right maxillary sinus (**Figure 3**). Endoscopy



Figure 3—Dorsoventral radiographic view of the skull of the horse in Figure 1 obtained during an examination performed in 2006 (2.5 years after resolution of initial clinical signs). Notice the abnormal soft tissue opacity within the right nasal cavity and right maxillary sinus.

revealed partial obstruction of the left nasal passage by a large mass located ventral to the ethmoid turbinates. In the right nasal passage, the presence of multiple coalescing nodules (1 to 3 cm in diameter) resulted in occlusion, preventing passage of the endoscope beyond a point 15 cm from the nares. A uterine biopsy instrument was used to obtain specimens from a rostrally located mass (10 cm from the nares) in the right nasal passage. Profuse postbiopsy hemorrhage was resolved following topical administration of 15 mL of 1% phenylephrine hydrochloride nasal spray.[g] Injection of amphotericin B[f] into the masses was unsuccessful because of hemorrhage. Thoracic radiography revealed no abnormalities.

Findings of cytologic and histologic examinations of the biopsy samples were inconclusive; only granulation tissue and evidence of chronic inflammation were observed. Tissue samples were submitted to 2 reference laboratories for latex agglutination *Cryptococcus* antigen testing, and results of both tests were positive (1:40 at the University of Georgia and 1:15 at Colorado State University). Results of serologic testing (agar gel immunodiffusion) performed at Colorado State University indicated that the horse did not have blastomycosis, coccidiodomycosis, or histoplasmosis.

Prior to receiving those test results, antifungal treatment was initiated based on the previous diagnosis of cryptococcal infection. Fluconazole[h] was administered orally as a 14 mg/kg loading dose, followed by a dosage of 5 mg/kg, PO, every 24 hours for 8 months (discontinuation of treatment was dependent on clinical response and findings at recheck examinations), and amphotericin B (93 μg/kg [42.4 μg/lb]) was administered IV every 8 hours for 7 days.[4] The amphotericin B was diluted in 1 L of 5% dextrose solution and delivered over a period of approximately 60 minutes. One hour prior to each amphotericin B infusion, the horse was administered 2 tablespoons (approx 30 g) of sodium chloride (table salt) orally as a slurry with corn syrup to decrease nephrotoxic effects.[5] Flunixin meglumine (500 mg, PO, q 24 h) was administered for 2 days. Serum creatinine concentration was monitored every 3 days, and values remained within the reference interval. Urine specific gravity determination and sediment examination were performed every 2 to 3 days; urine specific gravity remained between 1.025 and 1.035, and no casts were observed. Nasal discharge improved slightly, and after 7 days, the exophthalmos was noticeably decreased. The dosage of amphotericin B was increased (156 μg/kg [70.9 μg/lb], IV, q 8 h) for 7 days. During the 2-week period, the total amount of amphotericin B administered was 2.8 g.

Endoscopy was repeated 12 days after the horse was admitted to the hospital. At this time, it was possible to pass the endoscope through the right nasal passage. Several coalescing masses were located along the length of the nasal passage. One of the masses on the right side was wrapped around the caudal aspect of the nasal septum and was visible during endoscopy of the left nasal passage. After 14 days of amphotericin B and fluconazole treatment, results of a CBC and serum biochemical analyses were within reference limits except for mild anemia (PCV, 29.6%), decrease in the globulin concentration (5.4 g/dL), and mild hyperfibrinogenemia (500 mg/dL; reference interval, 100 to 400 mg/dL).

EQUINE

The horse was anesthetized, and surgical debulking of the masses was performed. Access to the right conchofrontal, maxillary, and ventral conchal sinuses was obtained via a large (6 × 15-cm) frontonasal sinusotomy bone flap as described previously.[6] A gelatinous, cystic mass that filled the sinus cavity was debulked via blunt dissection. An opening was made through the ventral conchal sinus into the right nasal passage for removal of masses from the nasopharynx. Complete removal of abnormal tissue was limited by access and hemorrhage. The blood loss volume during the procedure was approximately 13 L, and the horse received 8 L of a balanced electrolyte solution and 400 mL of 23% calcium gluconate during surgery. After removal of the accessible masses, the sinus cavity was packed with rolled gauze soaked in iced saline (0.9% NaCl) solution. The periostium of the bone flap was closed by use of 0 polydioxinone suture and the skin closed with skin staples. An 18-F red rubber catheter was inserted at a separate entry point into the sinus to facilitate postoperative lavage. An indwelling nasotracheal tube was placed in the left nostril to maintain a patent airway, and supplemental oxygen insufflation (10 L/min) was provided during recovery from anesthesia.

After recovery from anesthesia, the horse received 4 L of fresh, cross-matched whole blood that had been collected from a gelding from the Auburn University blood donor herd prior to surgery (administered over a 2-hour period) and 30 L of a balanced electrolyte solution (administered over a 12-hour period). Penicillin G potassium (22,000 U/kg [10,000 U/lb], IV, q 6 h for 3 days) and phenylbutazone (2 mg/kg [0.9 mg/lb], PO, q 12 h for 3 days, then q 24 h for 5 days) were administered after surgery. The sinus was lavaged daily with sterile saline solution. The packing was removed from the sinus 4 days after surgery. Five days after surgery, endoscopic examination of the nasal passages revealed a large blood clot obstructing the right nasal passage and a remaining mass near the left ethmoid turbinate.

The horse developed a localized infection around 2 of the staples, which were subsequently removed. Trimethoprim sulfonamide (20 mg/kg [9 mg/lb] PO, q 12 h) was administered for 10 days. The horse was discharged from the hospital, and the owners were instructed to continue treatments with fluconazole and trimethoprim sulfonamide.

Cytologic and histologic examination of tissues removed during surgery revealed organisms that were consistent with *Cryptococcus* spp. Histologically, there was evidence of chronic granulomatous sinusitis with free and phagocytosed basophilic organisms (3 to 7 μm in diameter) that were surrounded by a capsule that stained with Mayer's mucicarmine stain. There was no evidence of ethmoidal hematoma within the removed tissue. Fungal culture of tissue specimens yielded growth of *Cryptococcus neoformans*.

The horse was reexamined 6 weeks after undergoing surgical debulking of the nasopharyngeal masses. The severity of stertorous breathing and mucopurulent discharge from the right nostril and right medial canthus was decreased, compared with findings during the period of hospitalization prior to surgery, but was still intermittently detectable. Endoscopic examination of the nasal passages and nasopharynx revealed that the remaining lesions were approximately half the size that they were at the time of discharge from the hospital following surgery.

Following fungal culture of excised tissue samples, results of antimicrobial susceptibility testing indicated that the organism was susceptible to clotrimazole, ketoconazole, miconazole, amphotericin B, nystatin, and itraconazole and had intermediate susceptibility to fluconazole. Treatment with itraconazole was considered, but the commercially available solution[i] of the drug was prohibitively expensive. Because of the lack of other feasible drug protocols and the apparent response to treatment, administration of fluconazole was continued.

The horse was reexamined in January 2007 to ensure that no additional lesions had developed. The owners stated that the horse occasionally had episodes of head shaking and nose scratching. Mild respiratory stridor was heard during exercise. Findings of a physical examination were within reference limits, and the horse weighed 539 kg (1,186 lb). Upper airway endoscopic examination revealed that both nasal passages appeared normal with the exception of a small mass (0.7 cm in diameter) observed on the right dorsal aspect of the nasopharynx (**Figure 4**). The large surgically created opening in the nasal conchae allowed observation of the single sinus cavity created during the previous surgery. No masses were evident in the sinus cavity. Cytologic examination and fungal culture of a small piece of tissue collected by use of endoscopic biopsy forceps failed to reveal any cryptococcal organisms. The serum *Cryptococcus* antigen titer (performed at the University of Georgia) was reduced from 1:40 (determined in July 2006) to 1:4. Fluconazole treatment was continued for an additional month (8 months of treatment in total). Endoscopic reexamination was advised every 4 to 6 months; however, this was not performed until 16 months later.

In May 2008, the horse was reexamined at the teaching hospital to ensure there was no recurrence of fungal infection. Physical examination findings were
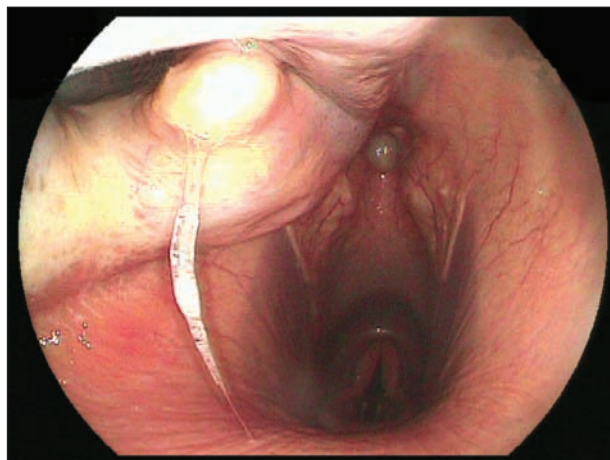


Figure 4—Endoscopic view of the larynx of the horse in Figure 1 obtained approximately 8 months after surgical debulking of mycotic lesions in the nasal passages performed to treat recurrence of cryptococcosis. One remaining mass is visible on the right dorsal aspect of the nasopharynx at the level of the surgically created sinusotomy.

within reference limits. The horse had a body condition score of 7 (scale of 9) and weighed 582 kg (1,280 lb). A stertorous inspiratory sound was heard during trotting exercise and sedation. Results of a CBC, fibrinogen concentration assay, and serum biochemical analyses were unremarkable. Skull radiography revealed thickening of the nasal septum, but no other abnormalities. Upper airway endoscopy revealed a narrowed right nasal passage. The small mass previously observed in the right dorsal aspect of the nasopharynx was reduced in size to a small raised area (0.3 cm in diameter), which was considered residual scar tissue. Biopsy samples of this raised area were collected, but cytologic examination and fungal culture of the specimens did not reveal evidence of fungal disease. The large right-sided sinus cavity created during the initial surgery was lined with apparently normal respiratory tract epithelium. A small amount of feed material and minimal mucopurulent discharge were observed in the cavity. Serum samples underwent cryptococcal antigen titer determination at the University of Georgia; the test result was 1:2 and considered negative. Results of all diagnostic procedures were indicative of resolution of the cryptococcal infection.

## Discussion

Although cryptococcal infection of the nasal cavity and sinuses in horses has been reported,[7–14] to our knowledge, this is the first case report of successful treatment of sinonasal cryptococcosis in a horse. Cryptococcosis has been successfully treated in 2 horses; 1 had a jejunal lesion with secondary intussusception that was surgically corrected,[15] and 1 had pneumonia that was treated IV with amphotericin B.[4] *Cryptococcus* spp have a predilection for the respiratory tract and CNS, and in horses, the organisms have been associated with pneumonia,[4,10,16–19] rhinitis,[7–11,13,14,20] sinusitis,[12] meningitis,[18,21–26] intestinal tract obstruction,[15] endometritis,[27] and abortion.[17,27] Of 41 cases of cryptococcosis in horses in the veterinary medical literature, 39 (95%) were fatal.[4,8–30] Treatments have included unsuccessful attempts at surgical removal,[28] cryotherapy,[20] topical administration of amphotericin B and nystatin,[11] and parenteral treatment with sodium iodide[18,28] and amphotericin B.[4,11,20,25]

Cryptococcosis is caused by *C neoformans* var *grubii* (serotype A), *C neoformans* var *neoformans* (serotype D), and *Cryptococcus gattii* (serotypes B and C).[31] Cryptococcal organisms are ubiquitous, saprophytic, round, basidiomycetous yeast-like fungi (5 to 10 μm in diameter); they have a large heteropolysaccharide capsule (1 to 30 μm in thickness) that does not react with common cytologic stains.[1,27] Histologically, cryptococcal lesions are characterized as having a soap bubble appearance because of the unstained capsules of massed organisms.[1] Cryptococcal species are the only pathogenic fungi that have a capsule. The capsule is easily identified with Mayer's mucicarmine stain, whereas the yeast bodies stain with periodic acid–Schiff and methenamine silver stains.[1] The inflammatory reaction to cryptococcal organisms is usually minimal, involving few macrophages, lymphocytes, and plasma cells. Granulomatous lesions are typified by numerous epithelioid macrophages and some multinucleated giant cells, but the severity of these lesions is often quite minimal, compared with lesions associated with other mycotic infections.[1] Cytologic or histologic identification of thickly encapsulated coccoid budding yeast bodies is considered sufficient to distinguish *Cryptococcus* spp from other fungi that also cause granulomatous upper respiratory tract lesions in horses, such as *Coccidioides immitis*, *Conidiobolus coronatus*, *Pseudallescheria boydii,* and *Rhinosporidium seeberi*.[1,14] In the horse of this report, a presumptive diagnosis was made in 2003 on the basis of examination of modified Wright-stained impression smears and H&E-stained tissue sections. For definitive diagnosis and differentiation among varieties of cryptococci, microbial culture or immunohistochemical staining is required. In 2006, the diagnosis of cryptococcosis was made on the basis of characteristic cytologic and histopathologic findings (including staining of the fungal capsule with Mayer's mucicarmine stain) in tissues that had been removed during surgery. The etiologic agent was confirmed on the basis of results of fungal culture and serologic evaluation. Colonies that grow on Sabouraud or inhibitory mold agar can be identified by use of a commercially available yeast identification kit.[27] Immunohistochemical techniques involving monoclonal antibodies are required to distinguish between serotypes *C neoformans* var *grubii* and *C neoformans* var *neoformans*; the diagnosis of *C gattii* infection is based on exclusion of the other cryptococcal varieties. Immunohistochemical staining was not performed as part of the diagnostic procedures for the horse of this report because the results would not have changed the treatment regimen. Discrepancies between immunohistochemical findings and results of fungal culture and serologic testing have been reported, and thus, the latter 2 techniques are still considered gold standards for identification of *Cryptococcus* spp.[32] Latex agglutination testing for identification of cryptococcal capsular antigen in serum is useful in the diagnosis of externally inapparent cryptococcal infections and determination of the specific species or variant of infective *C neoformans*. In a study[32] of a cryptococcal outbreak in cats, dogs, and ferrets in British Columbia, results indicated that the sensitivity and specificity of serologic testing for diagnosis of cryptococcosis are 92% and 95%, respectively. Disease resolution has been correlated with decreasing titers of *Cryptococcus* antigen in serum.[4,32]

In the environment, cryptococci are not encapsulated, but within tissues, the gelatinous capsule inhibits opsonic antibodies and protects the yeast from phagocytosis. Cell-mediated immunity is important in preventing development of disease. In people, HIV infection is an important predisposing factor in approximately 80% to 90% of cryptococcal infections.[33] In the absence of HIV infection, several conditions, including lymphoma, chronic leukemia, collagen vascular disease, sarcoidosis, cancer, immunosuppressive drug treatments, and diabetes mellitus, are often identified as underlying factors associated with cryptococcosis.[34,35] Ideally, immune function testing should have been performed in the horse of this report to identify any possible underlying immunodeficiency; however, the horse did not develop any other illnesses during the 6-year period.

EQUINE

Cryptococcosis has a worldwide distribution, but there appears to be a geographic predilection with relatively high frequency of infection in horses in Australia,[9,10,13,16,19,21,23] especially Western Australia[10,16,19]; recently, an outbreak of *C gattii* infection in British Columbia was reported.[32] There is an epidemiologic relationship between *C gattii* and the Australian river redgum tree (*Eucalyptus camaldulensis*),[19] whereas *C neoformans* var *neoformans* has historically been associated with bird (particularly pigeon) excreta. For the horse of this report, there was no known exposure to gum trees, poultry, or pigeons. Cryptococcal organisms are considered ubiquitous in the environment, and rare cases of cyptococcosis in horses in the southeastern United States have been reported.[j,k]

In the horse of this report, it was not possible initially to obtain a biopsy specimen of the mass in the upper portion of the right nasal passage to differentiate between ethmoidal hematoma and fungal granuloma; however, the endoscopic and radiographic appearance and response to intralesional formalin treatment were consistent with a progressive ethmoidal hematoma. If the mass was an ethmoidal hematoma, it is uncertain whether it developed prior to or after development of the cryptococcal granulomas. Development of an ethmoidal hematoma may have preceded development of the fungal granulomas, and the bloody discharge may have created a suitable environment for colonization by the yeast. In 1 report,[19] evidence of exercise-induced pulmonary hemorrhage was detected at necropsy in 3 of 7 horses that died as a result of cryptococcosis. The authors of that report suggested that pulmonary hemorrhage might have predisposed colonization of the lungs by inhaled cryptococcal organisms.[19] In another case report[13] of nasal cryptococcosis in a horse, a possible relationship between development of disease and a previous episode of severe traumatic epistaxis was suggested. Surgically resected tissues from the horse of this report (obtained in 2006) did not provide evidence of an ethmoidal hematoma; thus, either the ethmoidal hematoma had resolved in response to repeated intralesional formalin injections or all masses present in the horse's nasal passages in 2003 were fungal granulomas.

Amphotericin B has been used for the treatment of cryptococcosis in humans, dogs, and cats; however, it is nephrotoxic and irritates endothelial cells, often resulting in phlebitis. A pony with multiple pulmonary cryptococcal granulomas was successfully treated with daily IV infusions of amphotericin B over a 1-month period.[4] However, a 4-week period of IV treatment with amphotericin B was unsuccessful in a horse with cryptococcal meningitis.[25] In cats and people, administration of 5-flucytosine is an effective alternative to treatment with amphotericin B. Unfortunately, this drug is more expensive than amphotericin B and has been associated with comparatively higher incidence of fungal resistance and more frequent relapses.[36] Ketoconazole alone or in combination with 5-flucytosine has been used successfully to treat *Cryptococcus* spp infections in cats.[36] Ketoconazole is seldom used to treat horses because it requires an acid pH (acidification with 0.2N HCl) for its dissolution and gastrointestinal absorption and consequently has to be administered via nasogastric intubation.[37]

The horse of this report had cryptococcal granulomas of the upper respiratory tract, for which multimodal treatment was successful. Following the initial evaluation, the horse received oral administrations of fluconazole and adjunctive organic ethylenediamine dihydriodide and intralesional injections of fluconazole, amphotericin B, and formalin for a period of 6 weeks. Following recurrence of the disease, the horse underwent extensive surgical debulking of the lesions and received amphotericin B IV (2 weeks) and fluconazole orally (8 months).

Fluconazole is well absorbed after oral administration in horses. It is water soluble, minimally protein bound, distributes well in body tissues and fluids, and has a wide margin of safety.[2] In horses, a loading dose (14 mg/kg, PO) followed by administration of 5 mg of fluconazole/kg every 24 hours yields concentrations in plasma, CSF, synovial fluid, aqueous humor, and urine that are considered therapeutic against many fungal pathogens.[2] Fluconazole is especially useful for the treatment of coccidiodomycosis, blastomycosis, and histoplasmosis in humans[2] and has been used to successfully treat 2 horses with pulmonary coccidioidomycosis[38] as well as 2 pregnant mares with nasal conidiobolomycosis.[39]

The exact mode of action of iodides against fungal infections is unknown—they have very little, if any, direct in vitro antimicrobial effect.[3] Although iodides have been used as primary or adjunctive treatment of fungal disease in horses,[40] the overall efficacy is considered limited at best. Treatment is inexpensive, but toxicoses (characterized by excessive lacrimation, nonproductive cough, increased respiratory tract secretions, and dermatitis) can develop.[40] Additionally, administration of iodine in the diet of pregnant mares may cause congenital hypothyroidism in foals.[41]

In horses, mycotic granulomas have been successfully treated intralesionally with amphotericin B, and topical application of this antifungal agent was successful in the treatment of nasopharyngeal *Conidiobolus coronatus* infection.[42–45] In 2003, the owner of the horse of this report would not allow surgical debridement of the masses after severe hemorrhage occurred during biopsy procedures, and fluconazole was relatively expensive. Intralesional injection of formalin commonly is used to treat ethmoidal hematomas in horses.[46] Formalin injection of the fungal granulomas was considered a feasible and inexpensive means of chemical ablation.

The relative contributing effects of the multimodal treatment components to the overall success of the treatment in the horse of this report are unknown. Local treatment was performed easily at the times of reevaluation and may have decreased the duration and subsequent expense of orally administered antifungal treatment. However, despite amelioration of clinical signs for 2.5 years, the disease did recur. Unfortunately, veterinary attention was not sought by the owners until extensive disease developed. It is recommended that reevaluation of animals with known fungal infections be performed at least annually. Periodic endoscopy, radiography, and serologic testing for cryptococcal antigen may be beneficial.

In humans, combination antifungal regimens remain a mainstay in the treatment of systemic fungal infections.[47] A protocol consisting of a 2-week induction

phase with administration of either IV amphotericin B alone or in combination with oral 5-flucytosine, followed by a 10-week consolidation phase with administration of either oral fluconazole or oral itraconazole, has been advocated for the treatment of humans with cryptococcosis.[48–50] Alternative treatments include amphotericin B and 5-flucytosine administered for 6 to 10 weeks.[50]

Amphotericin B is a highly efficacious antifungal drug, but its expense prevented use of this drug in the horse of this report in 2003. Amphotericin B can be nephrotoxic, yet in humans, severe nephrotoxicosis is uncommon and often reversible.[50] New liposomal formulations of amphotericin B dramatically reduce the risk of nephrotoxicosis.[51] The horse of this report was pretreated with table salt because sodium loading (either orally or IV) reduces the incidence and severity of amphotericin B–induced nephrotoxic effects in people.[5]

In 1 report,[4] severe C gattii pneumonia in a pony resolved following a 1-month regimen of IV administration of amphotericin B without detectable deleterious renal effects. Two weeks of amphotericin B treatment did not result in clinically apparent decrease in renal function in the horse of this report. However, surgical debulking of accessible lesions may be more cost-effective and efficacious than long-term administration of amphotericin B. For the owners of the horse of this report, the cost of 2 weeks of amphotericin B treatment, including 5% dextrose solution, infusion sets, and monitoring of renal function, was $1,700.

The availability of laboratories that perform antifungal susceptibility testing of specimens collected from horses with fungal diseases is limited, and in humans, results often do not correlate with clinical response to treatment.[52,53] For the horse of this report, results of antifungal susceptibility testing were obtained after 5 months of treatment with fluconazole. At that time, the lesions were almost completely resolved (determined via endoscopic assessment). The infective cryptococcal organisms had only intermediate susceptibility to fluconazole in vitro. However, fluconazole is known for low in vitro activity, compared with its in vivo effectiveness, which may be attributable to its excellent tissue solubility.[2] Treatment with fluconazole was continued for a total of 8 months because the orally administered alternative antifungal agents (itraconazole and voriconazole) were prohibitively expensive.

Although treatment of mycotic lesions with intralesional drug injections may be attempted, the recent availability of a generic fluconazole product has dramatically reduced the expense of long-term oral administration. The initial treatment given to the horse of this report involved oral administration of fluconazole and adjunctive intralesional drug injections for 6 weeks and oral administration of organic ethylenediamine dihydriodide for 4 weeks, which resulted in remission of clinical signs for 2.5 years. Results of serologic testing for cryptococcal antigen were correlated with disease resolution in the horse's second episode of cryptococcosis, and biannual reassessment has been recommended to the owners. In the second episode of disease, the lesions were much more extensive and the treatment was more aggressive. Surgical resection of mycotic granulomas is recommended whenever feasible. In the horse of this report, fluconazole administration was continued for a total of 8 months without any adverse effects. Although not performed, immune function testing is now available for horses and is probably warranted in cases of cryptococcal infection.

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