Disseminated mycotic infection caused by *Westerdykella* species in a German Shepherd Dog

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**Case Description**—A 5-year-old 34.3-kg (75.5-lb) neutered male German Shepherd Dog was evaluated because of chronic azotemia that was unresponsive to typical medical management.

**Clinical Findings**—Urinalysis revealed pyuria and fungal hyphae. Fungal culture of a urine sample grew a sterile mold that was identified as *Westerdykella* spp via PCR assay.

**Treatment and Outcome**—The dog was treated empirically with itraconazole orally and amphotericin B IV for 5 weeks. Because of progressive azotemia, treatment was modified to oral administration of posaconazole. The dog improved but then developed progressive azotemia, hyperphosphatemia, and suspected diskospondylitis. Treatment was again modified to oral administration of terbinafine on the basis of results of antifungal susceptibility testing. The dog was euthanized after 5 months of antifungal treatment because of a deteriorating clinical condition and progressive azotemia.

**Clinical Relevance**—*Westerdykella* spp are filamentous hyphal organisms from the family Sporomycetaceae and had not previously been reported to cause infections in dogs. Fungal PCR assay and antifungal susceptibility testing may be useful for a patient with a suspected fungal infection that does not respond to empirical treatment or when traditional culture methods for fungal identification are unsuccessful. *Westerdykella* spp should be considered as a possible etiologic agent when systemic mycosis is diagnosed. ([J Am Vet Med Assoc](https://www.jamva.org/article/S0361-6230(13)01977-0/abstract) 2013;242:381–387)

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A 5-year-old 34.3-kg (75.5-lb) neutered male German Shepherd Dog was evaluated at the University of Florida Small Animal Hospital because of chronic azotemia. Ten days previously, the referring veterinarian had examined the dog because of polyuria, polydipsia, and mild hyporexia. At that time, physical examination findings were unremarkable except for mild dehydration (approx % not provided in medical record). Results of a serum biochemical analysis revealed moderate azotemia (BUN concentration, 61 mg/dL [reference range, 7 to 25 mg/dL]; creatinine concentration, 4.4 mg/dL [reference range, 0.3 to 1.4 mg/dL]) and mild hyperglobulinemia (globulin concentration, 6.0 g/dL; reference range, 2.3 to 5.2 g/dL). Urinalysis revealed hyposthenuria (urine specific gravity, 1.007; reference range used unavailable) with no bacteruria. Thoracic radiographs revealed mild microcardia. Results of an ACTH stimulation test were not consistent with hypoadrenocorticism (pre-ACTH cortisol concentration, 3.7 µg/dL [reference range, 2 to 6 µg/dL]; post-ACTH cortisol concentration, 20.7 µg/dL [reference range, 6 to 18 µg/dL]). A PCR assay panel® to screen serum for vector-borne diseases yielded negative results for *Anaplasma phagocytophilum*, *Anaplasma platys*, *Babesia canis*, *Babesia gibsoni*, *Bartonella henselae*, *Bartonella vinsonii*, *Borellia burgdorferi*, *Ehrlichia* spp, *Mycoplasma hemocanis*, *Mycoplasma hematoparvum*, *Neorickettsia risticii*, and *Rickettsia rickettsii*. The dog was treated with isotonic crystalloid fluids® (27 mL/kg/d [12.3 mL/lb/d]), SC, ampicillin (14.6 mg/kg [6.6 mg/lb], PO, q 8 h), enrofloxacin (4.0 mg/kg [1.8 mg/lb], PO, q 12 h) and fed a commercial diet® formulated for dogs with renal disease. After 10 days of treatment, the dog’s condition had not improved, and the dog was referred to the small animal hospital at the University of Florida.

During initial evaluation at the small animal hospital, physical examination and CBC results were unremarkable. Results of a serum biochemical analysis revealed severe azotemia (BUN concentration, 93 mg/dL [reference range, 7 to 27 mg/dL]; creatinine concentration, 7.6 mg/dL [reference range, 0.8 to 1.7 mg/dL]) and mild hyperphosphatemia (phosphorus concentration, 7.6 mg/dL [reference range, 2.3 to 5.2 mg/dL]). Urinalysis of a urine sample obtained via cystocentesis revealed isosthenuria (urine specific gravity, 1.008), mild pyuria (8 to 12 WBCs/hpf; reference 0), and mild hematuria (5 to 10 RBCs/hpf; reference 0). Urinary sediment analysis revealed proteinuria (protein via sulfosalicylic acid precipitation, 1+; reference, 0), and urobilinogenuria (urobilinogen, 4+ to 5+; reference, 0). Results of a serum biochemical analysis revealed severe azotemia (BUN concentration, 93 mg/dL [reference range, 7 to 27 mg/dL]; creatinine concentration, 7.6 mg/dL [reference range, 0.8 to 1.7 mg/dL]) and mild hyperphosphatemia (phosphorus concentration, 7.6 mg/dL [reference range, 2.3 to 5.2 mg/dL]). Urinalysis of a urine sample obtained via cystocentesis revealed isosthenuria (urine specific gravity, 1.008), mild pyuria (8 to 12 WBCs/hpf; reference 0), and mild hematuria (5 to 10 RBCs/hpf; reference 0). Urinary sediment analysis revealed proteinuria (protein via sulfosalicylic acid precipitation, 1+; reference, 0), and urobilinogenuria (urobilinogen, 4+ to 5+; reference, 0).
range, 0 to 4 WBCs/hpf), and moderate numbers of fungal hyphae. Abdominal ultrasonography revealed bilateral loss of renal corticomedullary distinction. Thoracic radiographs were unremarkable. The dog was hospitalized and treated with IV fluids (lactated Ringer’s solution, 6 mL/kg/h [2.7 mL/lb/h]), doxycycline hyclate (4.3 mg/kg [2 mg/lb], PO, q 12 h), famotidine (1.7 mg/kg [0.77 mg/lb], PO, q 12 h), and aluminum hydroxide (24.5 mg/kg [11.1 mg/lb], PO, q 12 h). Because the dog was suspected of having systemic aspergillosis, treatment with itraconazole (5.8 mg/kg [2.6 mg/lb], PO, q 12 h) was initiated while fungal culture of a urine sample was being performed.

During the following week, the dog’s clinical condition deteriorated as evidenced by more severe lethargy and anorexia. The dog’s azotemia persisted, and moderate hyperphosphatemia (phosphorus concentration, 9.7 mg/dL) and mild hypoalbuminemia (albumin concentration, 2.5 g/dL; reference range, 2.9 to 3.7 g/dL) were detected. Liposome-encapsulated amphotericin B (1.5 mg/kg [0.7 mg/lb], IV, q 2 to 3 days; total cumulative dose, 12 to 24 mg/kg [5.3 to 10.9 mg/lb]) was added to the treatment regimen. Results of a microscopic agglutination test did not indicate a 4-fold increase in serum antibody titers against leptospirosis between an acute serum sample obtained during the initial examination by the referring veterinarian and a convalescent serum sample obtained 21 days later; therefore, doxycycline administration was discontinued. Eight days after initiation of amphotericin B treatment in addition to continued supportive care and itraconazole treatment, the dog’s clinical condition and serum biochemical values had improved (BUN concentration, 52 mg/dL; creatinine concentration, 4.7 mg/dL; phosphorus concentration, 4.7 mg/dL), except for persistent hypoalbuminemia (albumin concentration, 2.2 g/dL).

Two days later (18 days after initial examination at the University of Florida Small Animal Hospital hospital), the dog’s clinical condition continued to improve except it acutely developed signs of pain in the caudal cervical area. Radiographic results of the cervical spinal column and analysis of CSF obtained via cisternal puncture were unremarkable. Fungal diskospondylitis was suspected, and treatment with amphotericin B and itraconazole was continued. Additionally, analgesia was provided via initiation of treatment with methadone hydrochloride (0.1 mg/kg [0.05 mg/lb], IV, q 4 to 6 h), tramadol hydrochloride (2.3 mg/kg [1.05 mg/lb], PO, q 6 h), and gabapentin (6.25 mg/kg [2.84 mg/lb], PO, q 8 h). The signs of pain in the neck area decreased during the next 7 days.

Fungal culture of a urine sample yielded a mold (3,000 colonies/mL) that was sterile, which precluded fungal identification by traditional methods. Ultimately, DNA sequencing was necessary for fungal identification. Template preparation, PCR assay, and DNA sequencing were performed as described. The ITS and sequences of the D1/D2 region were then used to search GenBank, with ≥ 90% query coverage and ≥ 97% identity considered significant. The top 3 hits for the ITS search consisted of Pycnidio phora dispersa (GenBank accession No. AY943035.1), for which 533 of 533 (100% identity) bases matched; Westerdykella multispora (GenBank accession No. AY943048.1), for which 532 of 532 (100% identity) bases matched; and Phoma herbarum (GenBank accession No. DQ912692.1), for which 439 of 558 (79% identity) bases matched. The top 3 hits for the D1/D2 sequence search consisted of P dispersa (GenBank accession No. AY943035.1), for which 533 of 533 (100%) bases matched; W multispora (GenBank accession No. AY943048.1), for which 532 of 532 (100%) bases matched; and Pycnidio phora aurantiaca (GenBank accession No. AY943057.1), for which 511 of 523 (98%) bases matched. The top 2 hits from each search were Westerdykella dispersa (P dispersa is a synonym for W dispersa) and W multispora. Because of the 100% identity for both the ITS and D1/D2 sequence searches, ribosomal sequencing could not discriminate between these 2 species. Also, because distinguishing morphological characteristics were not produced in culture, the isolate was identified as a Westerdykella sp.

After 25 days of hospitalization and treatment administration, the dog’s condition had improved such that it no longer required IV fluids to maintain its hydration. The dog was discharged, and IV treatment with amphotericin B was continued on an outpatient basis. During subsequent visits to the hospital for amphotericin B treatment, the dog’s clinical status was monitored via serial serum biochemical analyses, urinalyses, and bacteriologic and fungal cultures of urine samples. Results of serum biochemical analyses revealed persistent and progressive worsening of azotemia, hyperphosphatemia, and hypoalbuminemia. Fungal organisms were consistently identified during urinalyses. Fluid therapy (saline [0.9% NaCl] solution; 33 mL/kg [15 mL/lb], SC, q 24 h) was initiated to maintain hydration. One week after initiation of fluid therapy, the dog continued to have moderately severe azotemia (BUN concentration, 102 mg/dL; creatinine concentration, 7.6 mg/dL), despite adequate hydration and no fungal hyphae observed on urinalysis. Nephrotoxicosis caused by amphotericin B was suspected, and administration of the drug was discontinued. Administration of itraconazole was also discontinued because of an apparent lack of efficacy, and posaconazole (5 mg/kg [2.3 mg/lb], PO, q 24 h) administration was initiated.

Seven days after initiation of posaconazole treatment, the dog’s azotemia had worsened (BUN concentration, 118 mg/dL; creatinine concentration, 8.3 mg/dL). Progression of mycotic nephritis was suspected, and the posaconazole dosage was increased (10 mg/kg [4.5 mg/lb/d], PO, divided, q 12 h on the basis of a report of increased bioavailability of posaconazole with
The dog’s azotemia improved slightly during the following 3 weeks. The dog was clinically stable, although it developed intermittent hyporexia, vomiting, and small bowel diarrhea, which were treated empirically with metoclopramide hydrochloride (0.2 mg/kg [0.09 mg/lb], PO, q 8 h), omeprazole (0.3 mg/kg [0.14 mg/lb], PO, q 12 h), and bismuth subsalicylate (262 mg, PO, q 12 h). During the following 3 weeks, the dog appeared comfortable; the hyporexia resolved, and the dog gained 1.36 kg (3 lb). Five weeks after the dog was discharged from the hospital, it was reevaluated because of a 3-day history of lethargy and pyrexia (41.7°C [107.1°F]; reference range, 37.5°C to 39.2°C [99.5°C to 102.5°C]). Physical examination findings were unremarkable except for signs of pain during palpation of the neck, lumbar spinal, and sternal areas. Thoracic radiographs revealed dynamic instability at sternebrae 5 and 6, which was suggestive of early diskospondylitis. Results of a serum biochemical analysis revealed an improvement of the azotemia (BUN concentration, 76 mg/dL; creatinine concentration, 7.3 mg/dL). Results of a CBC revealed a mild regenerative left shift categorized by a neutrophilic leukocytosis (WBC count, 29.94 × 10^3 WBCs/µL; neutrophil count, 28 × 10^3 neutrophils/µL [reference range, 3.0 × 10^3 to 14.5 × 10^3 WBCs/µL]; neutrophil count, 28 × 10^3 neutrophils/µL [reference range, 3.0 × 10^3 neutrophils/µL to 11.5 × 10^3 neutrophils/µL]; and band neutrophil count, 0.9 × 10^3 band neutrophils/µL [reference range, 0 × 10^3 band neutrophils/µL to 0.3 × 10^3 band neutrophils/µL]). Cytologic evaluation of a fine-needle aspirate of the soft tissue surrounding the fifth sternebrae revealed minimal neutrophilic inflammation. Bacteriologic and fungal culture of serial urine and blood samples were performed and yielded no bacterial or fungal growth. The dog was hospitalized and treated with IV fluid therapy (saline solution; 4.8 mL/kg/h [2.2 mL/lb/h]) for medical management of chronic kidney disease and a combination of ampicillin sodium and sulfactam sodium (22 mg/kg [10 mg/lb], IV, q 8 h) for possible sepsis. Within 24 hours after hospitalization, the dog became normothermic and the lethargy resolved. The dog was discharged after 3 days of hospitalization with the addition of a 2-week prescription of amoxicillin-clavulanic acid (16.8 mg/kg [7.6 mg/lb], PO, q 12 h) to its previous medical treatment regimen (posaconazole, metoclopramide, omeprazole, bismuth subsalicylate, and gabapentin). During the following week, the dog remained afebrile and no longer had signs of pain when the sternum was palpated. The dog continued to have signs of mild discomfort during palpation of the thoracolumbar spinous region, but radiographs did not reveal any evidence of diskospondylitis in that area. Necropsy findings included marked tubulointerstitial nephritis and pyelonephritis with moderate hydrenephrosis, mild myocardial fibrosis, moderate osteosclerosis of the thoracic vertebrae, and necrotizing osteochondritis in the fourth through sixth sternebrae. Histopathologic findings included marked chronic nonsuppurative fibrosing interstitial nephritis and pyelonephritis with the presence of intrapelvic and intralesional fungal hyphae as well as moderate atherosclerosis with intralesional fungal hyphae and moderate hydronephrosis (Figures 2 and 3), necrotizing osteochondritis of the sternebrae with intralesional fungal hyphae (Figure 4), moderate osteosclerosis and periosteal ossification of the fourth through eighth sternebrae.

Figure 1—Lateral thoracic radiographic view of a 5-year-old German Shepherd Dog with a disseminated mycotic infection caused by Westerdykella spp. Notice the lytic lesions at the intersternebral space between the fourth and fifth sternebrae and between the fifth and sixth sternebrae (arrows) and the more subtle lytic lesions at intervertebral spaces T4-5, T5-6, T9-10, and T10-11.
thoracic vertebrae, mild histiocytic inflammation with moderate myocardial fibrosis, and a fibrin thrombus with fungal hyphae and atherosclerotic vessels within the pancreas.

Discussion

To our knowledge, the present report is the first to describe an infection caused by *Westerdykella* spp in a dog. Disseminated mycoses are infrequently reported in dogs. In a retrospective study that evaluated the medical records of dogs admitted to a veterinary teaching hospital between January 1990 and March 2007, dogs with systemic aspergillosis represented only 0.02% of all dogs admitted to the hospital. However, 20 of the 30 dogs with systemic aspergillosis in that study were German Shepherd Dogs, and those 20 dogs represented 4.5% of all German Shepherd Dogs examined at that hospital. German Shepherd Dogs appear to have a genetic predisposition for the development of disseminated aspergillosis that may be related to an IgA deficiency. The most commonly reported organism isolated from dogs with disseminated mycoses is *Aspergillus terreus*. Other hyphal mycotic infections such as those caused by *Geosmithia*, *Penicillium*, *Paecilomyces*, *Sagenomella*, *Adiaspiromyces*, *Phaeohyphomycoses*, and *Oxyporus* spp have also been reported in German Shepherd Dogs.

*Westerdykella multispora*, an ascomycete, is a filamentous hyphal organism from the family Sporomycaceae. Available literature regarding *W multispora* describes dykell acid, a novel metabolite of the organism, that is suspected to inhibit drug-induced apoptosis via a caspase-3–like protease-suppressing mechanism and is also thought to have antimetastatic and antiangiogenic activity. *Westerdykella dispersa* is reported to produce the antifungal agents lanomycin and glucolanomycin. To our knowledge, *Westerdykella* spp have not previously been reported to be pathogenic.

In the present report, the dog with disseminated mycosis caused by *Westerdykella* spp had clinical signs...
associated with acute renal failure and diskospondylitis that were similar to those reported in dogs with disseminated aspergillosis. The dog of the present report was empirically treated in accordance with current treatment recommendations for disseminated aspergillosis until results of molecular diagnostic testing revealed that the pathogen was not an *Aspergillus* sp or other mycotic infection for which treatment recommendations were available. This prompted additional antifungal susceptibility testing in an attempt to determine an appropriate treatment because the *Westerdykella* organism was not susceptible to itraconazole, the antifungal agent commonly used to treat disseminated aspergillosis, in vitro or in vivo.

Fungal culture is the gold standard for diagnosis of a mycotic infection. Fungal identification requires evaluation of both gross colony morphology and microscopic examination of the reproductive structures of the organism. Unfortunately, fungal culture may be unsuccessful because of several factors, including a low number of fungal organisms in a sample, lack of fungal growth, and the use of inappropriate media for fungal growth. Sporulation is required for fungal identification because examination of the fruiting bodies, conidia, or arthrospores is necessary to complete the fungal description. The fungus cultured from the dog of the present report grew a sterile mold that would not sporulate despite numerous efforts, which precluded identification by traditional fungal culture methods. As a result, other diagnostic methods such as an immunochemical assay, *Aspergillus* galactomannan assay, and molecular diagnostic tests were considered.

Molecular diagnostic tests used for fungal identification include DNA sequencing of isolated organisms as well as a panfungal PCR assay. The advantages of PCR-based molecular diagnostic tests are that they do not require fungal sporulation and are very sensitive and often quicker than fungal culture. For the dog of the present report, a panfungal PCR assay and DNA sequencing were performed on the mold grown from a urine sample, which allowed the infecting organism to be identified as *Westerdykella* spp. The prevalence of fungemia in dogs with disseminated mycoses is unknown; however, in human patients, the use of a panfungal PCR assay to screen blood samples for fungal organisms is recognized as an opportunity for early diagnosis of disseminated hyphal mycoses. Panfungal PCR assay and DNA sequencing may be valuable tools for identification of fungal organisms that cannot be distinguished by traditional culture methods. Once fungal identification is made, empirical treatment can be selected on the basis of reported antifungal efficacy when available.

Antifungal susceptibility testing to determine the MICs for various antifungal agents against a particular fungal organism can help determine whether appropriate empirical antifungal treatment has been selected, which is important because patients with mycotic infections often require treatment with antifungal drugs for many months to years. Antifungal susceptibility testing also allows for selection of the least costly effective antifungal treatment. However, several limitations preclude the routine use of antifungal susceptibility testing, including limited availability, cost, and the amount of time it takes for results to become available. Reference ranges for MICs of antifungal drugs used in dogs have not been established; therefore, antifungal treatment recommendations are generally based on in vitro assessment, in which the antifungal drug with the lowest MIC is considered to be the most effective against the fungal pathogen in question. An inherent limitation of antifungal susceptibility testing is that the MICs are determined on the basis of antifungal drug concentrations in plasma rather than tissue, and information regarding concentrations of antifungal drugs achievable in tissue in vivo is often lacking. Investigators of other reports suggest that the ratio of the AUC to the MIC can be used to predict the efficacy of an antifungal drug in vivo and the optimal AUC:MIC ratio for posaconazole is 25:1. For a dog administered posaconazole (10 mg/kg, PO), the AUC for posaconazole is 26.0 µg·h/mL. The MIC for posaconazole against the *Westerdykella* spp pathogen infecting the dog of the present report was 0.125 µg/mL; therefore, the AUC:MIC ratio was 212.8:1, which exceeded the optimal AUC:MIC ratio. For the dog of the present report, initial treatment with itraconazole was clinically ineffective, and results of antifungal susceptibility testing indicated that itraconazole had an MIC > 4 µg/mL against the *Westerdykella* spp pathogen. Therefore, treatment was changed from itraconazole to posaconazole on the basis of results of the antifungal susceptibility testing. Later, treatment was changed from posaconazole to terbinafine because of financial constraints as well as worsening of azotemia, which was thought to be caused by the development of resistance to posaconazole. The optimal AUC:MIC ratio for terbinafine ranges from 9:1 to 13:1 for various systemic mycoses, such as Blastomyces dermatitidis, Histoplasma capsulatum, Sporothrix schenckii, and Coccidioides immitis. Oral administration of terbinafine (30 to 35 mg/kg) results in an AUC for terbinafine of 15.4 to 17.25 µg·h/mL. Dose recommendations for terbinafine vary depending on the organism being treated. The dog of the present report was treated with the dosage of terbinafine recommended for the treatment of systemic aspergillosis (5 to 10 mg/kg, PO, q 12 h), which is lower than the dosage of terbinafine recommended for the treatment of dermatophytosis (30 mg/kg [13.6 mg/lb], PO, q 24 h). To our knowledge, the AUC for terbinafine after being administered at a dose of 8 mg/kg, PO, has not been reported; thus, the AUC:MIC ratio for terbinafine in the present report could not be calculated. Despite the fact that the results of antifungal susceptibility testing suggested that terbinafine should be effective against the *Westerdykella* spp pathogen, the dog's clinical condition continued to decline after initiation of terbinafine treatment.

In addition to documenting a novel systemic mycosis in a dog, the present case report highlights many of the difficulties encountered in the treatment of a patient with disseminated mycosis. Most antifungal drugs, including the azoles and allylamines, are not fungicidal at systemically achievable concentrations; therefore, elimination of a mycotic infection is unlikely unless the patient has a competent immune system or the source of infection can be surgically removed. Generally, patients with systemic mycoses are suspected to...
have some impairment of the immune system, which makes treatment with fungicidal drugs such as amphotericin B desirable. For the dog of the present report, progressive renal dysfunction did not allow for continued treatment with amphotericin B.

When the dog of the present report appeared to develop resistance to posaconazole, antifungal treatment was changed to terbinafine on the basis of results of antifungal susceptibility testing; however, the dog’s clinical condition continued to worsen. There are multiple possible reasons for failure of terbinafine treatment. To our knowledge, concentrations of terbinafine biologically achievable in tissues such as kidney and bone have not been reported; therefore, it is possible that the dose of terbinafine administered to the dog of the present report was insufficient to adequately penetrate the tissues infected with the *Westerdykella* spp. Most reports regarding the administration of terbinafine to dogs involve the treatment of dermatophytosis, and the doses of terbinafine used were higher than that used for the dog of the present report. Studies evaluating the effectiveness of terbinafine for the treatment of systemic mycoses in species of veterinary interest are lacking. It is also possible that during the 3 months of posaconazole administration to the dog of the present report, the organism developed resistance to terbinafine as well as posaconazole. Terbinafine and posaconazole act at different phases of the ergosterol synthesis pathway, so alteration or overexpression of the enzyme targeted by posaconazole should not cause resistance to terbinafine. However, fungal organisms can also develop drug resistance by upregulation of efflux pumps that reduce the intracellular concentrations of antifungal drugs. This mechanism of resistance can affect azoles, such as posaconazole, as well as allylamines, such as terbinafine. In general, selection of an antifungal drug for the treatment of a patient with systemic mycosis should be guided by knowledge of the pharmacokinetics and pharmacodynamics of the antifungal drug in the species of interest, results of antifungal susceptibility testing, and any other mitigating factors, such as patients with concurrent immunosuppression.

The incidence of disseminated mycoses is increasing in human patients and possibly in dogs, most likely because of the use of immunosuppressive drugs and the increased frequency of diagnosis and treatment of hematologic and oncologic diseases. In dogs, disseminated mycosis is most frequently caused by *Aspergillus* spp but may also be caused by numerous other hyphal fungi. To our knowledge, disseminated mycosis caused by *Westerdykella* spp had not been described in either a human or veterinary patient prior to the present report. The use of molecular diagnostic methods can be used for accurate earlier diagnosis of mycotic infections, and antifungal susceptibility testing can be useful for the selection of an effective antifungal drug for the treatment of disseminated mycoses. However, because of the cost and other limitations of molecular diagnostic methods and antifungal susceptibility testing, it may be most practical to only pursue these methods when a patient with a suspected mycosis fails to respond to empirical antifungal treatment or when traditional culture methods for fungal identification are unsuccessful.

a. Idexx Laboratories, Brooksville, Fla.
b. Antech Diagnostics, Lake Success, NY.
e. Sparonox, Janssen-ortho, Toronto, ON, Canada.
g. Fungus Testing Laboratory, University of Texas Health Science Center, San Antonio, Tex.
h. Aspergillus galactomannan assay, Rosenstiel Medical, Miami, Fla.
i. Genbank, National Center for Biotechnology Information, Bethesda, Md.
j. Noxafl, Merck and Co Inc, Whitehouse Station, NJ.

References

Florfenicol concentrations in ovine tear fluid following intramuscular and subcutaneous administration and comparison with the minimum inhibitory concentrations against mycoplasmal strains potentially involved in infectious keratoconjunctivitis

Alain Regnier et al

**Objective**—To measure florfenicol concentrations in ovine tear fluid after IM and SC administration and determine minimum inhibitory concentrations (MICs) of florfenicol against field isolates of Mycoplasma organisms potentially involved in infectious keratoconjunctivitis.

**Animals**—9 healthy adult Lacaune ewes.

**Procedures**—Animals received an IM and SC administration of florfenicol (20 mg/kg) in a 2-way crossover design. Samples of blood and tear fluid were collected before and for 24 hours after administration. Concentrations of florfenicol in plasma and tear fluid were measured via high-performance liquid chromatography. The MIC of florfenicol for various Mycoplasma strains cultured from sheep and goats was determined via an agar dilution method.

**Results**—Mean florfenicol concentration in tear fluid for the 24-hour period was significantly higher after IM administration (0.70 µg/mL) than after SC administration (0.22 µg/mL) and was maintained for a longer duration. The lacrimal fluid-to-plasma concentration ratio was not different between the 2 routes of administration, with mean values of 40.2% and 32.5% after IM and SC administration, respectively. The MIC for Mycoplasma agalactiae, Mycoplasma conjunctivae, and Mycoplasma mycoides isolates ranged from 0.5 to 8 µg of florfenicol/mL. Two strains of M. agalactiae could be considered resistant to florfenicol.

**Conclusions and Clinical Relevance**—Florfenicol readily penetrated the precocular tear fluid of sheep after IM and SC administration. For both routes of administration, doses > 20 mg/kg would be necessary to achieve tear fluid concentrations of florfenicol greater than the MICs for most strains of Mycoplasma organisms. (Am J Vet Res 2013;74:268–274)