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# *Malassezia* spp on the periocular skin of dogs and their association with blepharitis, ocular discharge, and the application of ophthalmic medications

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**Objective**—To determine how frequently *Malassezia* spp were identified on the periocular skin of dogs and assess the respective associations between the presence of *Malassezia* spp on the periocular skin and blepharitis, ocular discharge, and the application of ophthalmic medications.

**Design**—Prospective clinical study.

**Animals**—167 eyelids of 84 dogs.

**Procedures**—Samples obtained from the surface of the eyelid skin by use of adhesive tape were evaluated cytologically for the presence of *Malassezia* spp. Dogs were grouped on the basis of the presence of blepharitis, nature of ocular discharge, and whether ophthalmic medications were applied, and the proportion of samples with *Malassezia* spp was compared among the groups.

**Results**—*Malassezia* spp were detected in 19 samples, of which 15 were obtained from eyes without blepharitis and 14 were obtained from eyes treated with topical ophthalmic medications. The proportion of samples with *Malassezia* spp was significantly higher for eyes with ocular discharge than for eyes without ocular discharge, especially if that discharge was mucoid or mucopurulent, and for eyes that were treated with aqueous-based medications only or a combination of oil- and aqueous-based medications than for eyes that were not treated.

**Conclusions and Clinical Relevance**—*Malassezia* organisms were detected on the periocular skin of 3 of 56 (5%) clinically normal dogs. *Malassezia* organisms were also frequently found on the periocular skin of dogs that had mucoid or mucopurulent ocular discharge or that were administered topical aqueous-based ophthalmic medications, and the periocular skin of these dogs should be cytologically evaluated for *Malassezia* organisms. (*J Am Vet Med Assoc* 2014;244:1304–1308)

*Malassezia* spp are lipophilic and often lipid-dependent yeasts that are common commensal organisms on the skin and ears of dogs but can also cause dermatologic and aural disease.<sup>1</sup> In veterinary medicine, one of the most clinically important species is *Malassezia pachydermatis*, which is an opportunistic pathogen commonly associated with canine dermatitis and otitis externa secondary to other diseases that result in abnormal sebaceous or ceruminous secretions. In dogs, the distribution of *Malassezia* spp is dependent on the location of skin sampled, disease status of the animal, and the method used for detection. *Malassezia* spp have been detected by cytologic evaluation in 40 of 99 (40%) healthy ears and 83 of 101 (82%) diseased ears,<sup>2</sup> and by culture of the organism from samples collected by the use of sterile cotton swabs moistened with sterile saline (0.9% NaCl)

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## ABBREVIATION

CI Confidence interval

solution to swab the skin of healthy dogs in various locations on their bodies.<sup>3</sup> In the study<sup>3</sup> that used culture as a means to identify *Malassezia* spp, the perianal area was the area most frequently colonized (20/33 [61%]), the inguinal area was the area least frequently colonized (1/33 [3%]), and *Malassezia* spp were cultured from the periorbital region of 3 of the 33 (9%) healthy study dogs. To our knowledge, studies have not been conducted in which the frequency of *Malassezia* colonization of the periocular region of healthy dogs was determined by cytologic examination. Furthermore, studies to investigate the association of *Malassezia* spp with blepharitis or periocular dermatitis are lacking but are warranted given that topical application of oil-based ophthalmic medications may affect the lipid composition of the periocular dermal microenvironment, which could promote the growth of *Malassezia* spp on the eyelid skin.<sup>4</sup> Additionally, many commonly used topical ophthalmic medications contain immunosuppressive agents such as corticosteroids, cyclosporine, or tacrolimus that might alter the cutaneous immunity and microflora of the periocular region. *Malassezia* spp can induce inflammation by either nonspecific mechanisms or antigen-specific hyper-

sensitivity reactions.<sup>5</sup> Thus, proliferation of *Malassezia* organisms in the periocular region could cause changes in the skin and localized dermatitis or blepharitis, particularly in patients that are hypersensitive to *Malassezia*. The purpose of the study reported here was to determine how frequently *Malassezia* spp were identified on the periocular skin of dogs with or without blepharitis and to evaluate the respective associations between the presence of *Malassezia* spp on the periocular skin and the presence of blepharitis, the presence and type of ocular discharge, and the topical application of oil-based, aqueous-based, or potentially immunosuppressive ophthalmic medications.

## Materials and Methods

**Animals**—Dogs examined by the Ophthalmology, Dermatology, or Community Medicine Services at the University of California-Davis William R. Pritchard Veterinary Medical Teaching Hospital from June 2012 through January 2013 were eligible for inclusion in the study and were enrolled on the basis of investigator availability and owner consent. Dogs were eligible for enrollment regardless of whether they were being treated with topical ophthalmic medications; however, dogs in which topical ophthalmic treatment had been discontinued < 3 days prior to examination or initiated < 7 days prior to examination were excluded from the study. Dogs were also excluded from the study if they had painful ophthalmic conditions or fragile globes subsequent to deep stromal ulcers, keratomalacia, corneal lacerations, recent intraocular surgery, acute glaucoma, anterior lens luxation, or traumatic proptosis. The study was approved by the hospital's Clinical Trials Review Board, and written consent was obtained from the owner of each dog prior to study enrollment.

**Sample collection and processing**—For each dog, clear cellophane adhesive tape was used as described<sup>6,7</sup> to obtain a sample from the skin surface of the upper and lower eyelids of each eye for cytologic evaluation. Briefly, the upper and lower eyelids were held closed and a strip of adhesive tape was placed against the periocular skin of both eyelids simultaneously; gentle pressure was applied to the tape to ensure contact with the skin. Care was taken to ensure that the tape did not touch the cornea. The tape was then removed.

A drop of basophilic thiazine dye<sup>a</sup> solution was placed on a glass microscope slide, and then the tape containing the sample collected from the eyelid skin surface was adhered to the slide over the dye, which was dispersed by the application of digital pressure over the tape. All samples were examined with 1,000 $\times$  magnification by a trained observer (GMN) who was unaware of the clinical history and signalment of the dogs from which the samples were obtained. For each sample, the number of *Malassezia* organisms was counted in 10 hpfs that contained keratinocytes or surface debris, and the mean number of *Malassezia* organisms/hpf was determined. At least 15 hpfs from each quadrant of the slide (ie, at least 60 hpfs) were examined before a sample was classified as not containing *Malassezia* organisms.

**Data collection**—For each dog, historical and clinical data were obtained from an interview with the dog's

owner, clinical examination, and a thorough review of the medical record. Data collected included signalment; clinical status of periocular skin; nature of ocular discharge, which ophthalmic medications (if any) were topically administered; and the duration of treatment with those medications, when appropriate. For the purpose of the study reported here, blepharitis was defined as inflammation of the eyelid or immediate surrounding skin. Ocular discharge at the time of examination was classified as absent, periocular tear staining or epiphora, mucoid, or mucopurulent. Topically administered ophthalmic medications were classified as oil- or aqueous-based. Oil-based medications included those in which the vehicle was white petrolatum, lanolin, or coconut, corn, mineral, or olive oil. Aqueous-based medications included those in which the vehicle was sodium chloride, purified water, hydroxypropyl methylcellulose, hyaluronate, or hyaluronan. Medications that contained cyclosporine, tacrolimus, or glucocorticoids such as dexamethasone and prednisolone were classified as potentially immunosuppressive.

**Statistical analysis**—The exact  $\chi^2$  test was used to evaluate the independence of categorical variables and to contrast observed and expected contingency table cell frequencies. Outcomes of interest were the presence of *Malassezia* spp in the periocular sample and the presence of blepharitis, which were both dichotomous in nature. For each outcome, binary logistic regression with robust variance estimation to account for nonindependence when both eyes from the same dog were evaluated was used to assess its respective associations with potential determinants of outcomes.<sup>8</sup> Such determinants included age, sex (male or female), neutered status (neutered or not neutered), type of ocular discharge (absent, periocular tear staining or epiphora, mucoid, or mucopurulent), and types of medications administered (none; aqueous-based medications only, with no potentially immunosuppressive drugs; aqueous-based medications only that contained at least 1 potentially immunosuppressive drug; oil-based medications only, with no potentially immunosuppressive drugs; oil-based medications only that contained at least 1 potentially immunosuppressive drug; a combination of aqueous- and oil-based medications, with no potentially immunosuppressive drugs; or a combination of aqueous- and oil-based medications that contained at least 1 potentially immunosuppressive drug). For each analysis, the null hypothesis was that the OR = 1, and values of  $P < 0.05$  were considered significant. Results were reported as ORs and the corresponding 95% CIs and  $P$  values for those ORs. All analyses were performed with a commercially available software program.<sup>b</sup>

## Results

**Animals**—Eighty-four dogs were enrolled in the study, of which 30 were castrated males, 10 were sexually intact males, 37 were spayed females, and 7 were sexually intact females. The mean age for all dogs was 7 years (range, 0.25 to 15.4 years). The study dogs represented a diverse range of breeds, the most common of which were mixed breed ( $n = 21$ ), Cocker Spaniel (7), and Labrador Retriever and Golden Retriever (5 each).

Periocular samples were collected bilaterally in all dogs except a spayed female Shih Tzu that had 1 eye enucleated prior to study enrollment. Thus, 167 periocular samples were evaluated.

Descriptive data for periocular samples with or without *Malassezia* organisms that were obtained from eyes with or without blepharitis or ocular discharge were summarized (Table 1). The mean  $\pm$  SD age did not differ significantly between dogs in which *Malassezia* organisms were ( $7.2 \pm 3.9$  years) or were not ( $6.9 \pm 4.2$  years;  $P = 0.79$ ) detected in the periocular samples, between dogs with ( $5.8 \pm 3.1$  years) or without ( $7.0 \pm 4.3$  years;  $P = 0.18$ ) blepharitis, or between dogs with ( $8.1 \pm 4.5$  years) or without ( $6.5 \pm 4.0$  years;  $P = 0.091$ ) ocular discharge. Likewise, the sex distribution (without consideration of the neutered status) did not differ significantly between dogs in which *Malassezia* organisms were or were not detected in the periocular samples ( $P = 0.74$ ) or between dogs with or without ocular discharge ( $P = 0.046$ ); however, female dogs were significantly ( $P = 0.017$ ) more likely to have blepharitis than were male dogs (OR, 5.39; 95% CI, 1.35 to 21.60).

**Proportion of periocular samples in which *Malassezia* spp were detected**—*Malassezia* organisms were cytologically detected in 19 of 167 (11.4%) periocular samples, of which 15 were obtained from eyes that did not have blepharitis and 14 were obtained from eyes that were being treated with topical ophthalmic medications. The proportion of periocular samples with *Malassezia* spp did not differ significantly ( $P = 0.703$ ) between eyes with (4/28) and those without (15/139) blepharitis (OR, 1.38; 95% CI, 0.26 to 7.17). However, the proportion of periocular samples with *Malassezia* spp was significantly ( $P = 0.013$ ) higher for eyes with (11/45) ocular discharge, compared with that for eyes without ocular discharge (8/122; OR, 4.61; 95% CI, 1.38 to 15.42). Specifically, samples obtained from eyes with mucoid or mucopurulent discharge were significantly

( $P = 0.018$ ) more likely to contain *Malassezia* organisms than were samples obtained from eyes without ocular discharge (OR, 5.87; 95% CI, 1.38 to 25.37), whereas the proportion of samples with *Malassezia* spp did not differ significantly ( $P = 0.114$ ) between eyes with tear staining or epiphora and eyes without any ocular discharge (OR, 3.35; 95% CI, 0.75 to 15.06). The proportion of periocular samples with *Malassezia* spp did not differ significantly ( $P = 0.229$ ) between eyes that were (14/94) and those that were not (5/73) treated with topical ophthalmic medications (OR, 2.38; 95% CI, 0.58 to 9.77).

When the type of medication administered was considered, the proportion of periocular samples with *Malassezia* spp did not differ significantly ( $P = 0.422$ ) between eyes that received at least 1 oil-based medication (9/61) and those that received no (10/106) oil-based medications (OR, 1.66; 95% CI, 0.48 to 5.74); however, *Malassezia* organisms were detected more frequently in periocular samples collected from eyes that were treated with aqueous-based medications only (5/33), compared with those that were not treated with aqueous-based medications (5/102; OR, 5.33; 95% CI, 1.29 to 21.95;  $P = 0.021$ ). The proportion of samples with *Malassezia* spp did not differ significantly ( $P = 0.480$ ) between eyes treated with at least 1 potentially immunosuppressive drug (8/60) and those treated with no potentially immunosuppressive drugs (11/107; OR, 1.57; 95% CI, 0.45 to 5.44). *Malassezia* organisms were detected more frequently in periocular samples obtained from eyes treated with a combination of oil- and aqueous-based medications (9/32) than in samples obtained from eyes that were not treated with topical ophthalmic medications (5/73; OR, 4.89; 95% CI, 1.37 to 17.42;  $P = 0.014$ ). Contingency table analysis revealed that periocular samples obtained from eyes that were treated with aqueous-based medications that did not contain any immunosuppressive drugs and eyes that were treated with a combination of aqueous- and oil-based medications that contained at least 1 potentially immunosuppressive drug were significantly ( $P = 0.002$ ) more likely to contain *Malassezia* organisms than were samples obtained from eyes in all other medication groups. The number of topical ophthalmic medications administered to an eye was not significantly ( $P = 0.113$ ) associated with the presence of *Malassezia* spp in the periocular sample obtained from that eye. All periocular samples in which *Malassezia* organisms were detected were obtained from eyes that were treated with at least 1 of the following topical ophthalmic medications: 0.2%, 1%, or 2% compounded cyclosporine in corn or olive oil, 0.02% compounded tacrolimus in corn oil, tacrolimus drops (concentration unknown) in coconut oil, hyaluronate, hydroxypropyl methylcellulose-based artificial tears, ofloxacin solution, or prednisolone acetate suspension.

*Malassezia* organisms were detected in 16 of 111 (14%) periocular samples obtained from eyes with at least 1 of the 3 suspected risk factors assessed (blepharitis, ocular discharge, or administration of topical medication), whereas *Malassezia* organisms were detected in only 3 of 56 (5%) samples obtained from eyes without any of the suspected risk factors assessed. However, when considered collectively, the 3 suspected risk factors were not sig-

Table 1—Descriptive data for 84 dogs of various breeds from which 167 surface samples from the periocular skin were obtained by means of adhesive tape for cytologic evaluation for the presence of *Malassezia* spp.

Outcome	No. of eyes	Age (y)	No. of eyes from dogs of each sex	
			Male	Female
<i>Malassezia</i> organisms in periocular samples				
Present	19	7.9 (1.3–14.0)	10	9
Absent	148	7.1 (0.25–15.4)	70	78
Blepharitis				
Present	28	5.1 (1.2–12.0)	5	23*
Absent	139	7.2 (0.25–15.4)	75	64
Ocular discharge				
Present	45	8.5 (0.75–15.4)	14	31
Absent	122	6.1 (0.25–15.3)	66	56

Dogs were examined by the Ophthalmology, Dermatology, or Community Medicine Services at the University of California–Davis William R. Pritchard Veterinary Medical Teaching Hospital between June 2012 and January 2013. Surface samples from the periocular skin were obtained bilaterally from all dogs except for 1, which had an eye enucleated prior to study enrollment. Values for age represent the median (range).

\*Value differs significantly ( $P < 0.05$ ) from that for male dogs.



nificantly ( $P = 0.193$ ) associated with presence of *Malassezia* organisms (OR, 2.98; 95% CI, 0.58 to 15.36).

**Proportion of eyes with blepharitis**—Blepharitis was diagnosed in 28 of 167 (17%) eyes sampled. The majority (24/28) of samples from eyes with blepharitis had no detectable *Malassezia* organisms, and the majority (17/28) of dogs with blepharitis were not receiving ophthalmic medications. The proportion of samples obtained from eyes with blepharitis and from which *Malassezia* spp were detected (4/19) did not differ from the proportion of samples obtained from eyes with blepharitis and in which *Malassezia* spp were not detected (24/148; OR, 1.38; 95% CI, 0.26 to 7.17;  $P = 0.7$ ). However, the proportion of eyes with blepharitis and ocular discharge (15/45) was significantly ( $P = 0.015$ ) greater than the proportion of eyes with blepharitis and no ocular discharge (13/122; OR, 4.19; 95% CI, 1.32 to 13.33). Specifically, mucoid or mucopurulent discharge was significantly ( $P = 0.001$ ) associated with the presence of blepharitis (OR, 9.91; 95% CI, 2.66 to 36.94), whereas epiphora or tear staining was not (OR, 0.88; 95% CI, 0.097 to 8.01;  $P = 0.912$ ) associated with the presence of blepharitis. The proportion of eyes with blepharitis did not differ significantly ( $P = 0.156$ ) between eyes that were (17/94) and eyes that were not (11/73) treated with topical medications (OR, 0.44; 95% CI, 0.14 to 1.37).

When the type of medication administered was considered, the presence of blepharitis did not differ significantly between eyes that were treated with at least 1 oil-based medication (9/61) and those that were not treated with oil-based medications (19/106; OR, 0.79; 95% CI, 0.24 to 2.60;  $P = 0.702$ ), between eyes that were treated with aqueous-based medications only (2/33) and those that were not treated with aqueous-based medications (23/106; OR, 0.29; 95% CI, 0.07 to 1.15;  $P = 0.078$ ), between eyes that were treated with at least 1 potentially immunosuppressive drug (9/60) and those that were not treated with a potentially immunosuppressive drug (18/107; OR, 0.75; 95% CI, 0.23 to 2.46;  $P = 0.630$ ), or between eyes that were treated with a combination of oil- and aqueous-based medications (3/32) and those not treated with any topical medications (11/73; OR, 0.46; 95% CI, 0.09 to 2.42;  $P = 0.356$ ). The 11 eyes with blepharitis that were treated with topical medications were treated with at least one of the following: bacitracin-neomycin-polymyxin in mineral oil; white petrolatum; 0.2%, 1%, or 2% compounded cyclosporine in corn or olive oil; hyaluronate; or hydroxypropyl methylcellulose-based artificial tears.

## Discussion

To our knowledge, the present study is the first conducted in which cytologic evaluation was used to detect *Malassezia* spp on the periocular skin of dogs with or without evidence of ocular disease and the second to determine the frequency of detection of *Malassezia* spp on the periocular skin of dogs.<sup>3</sup> Results of other studies<sup>2,3</sup> indicate that the likelihood of detection of *Malassezia* spp on the skin of dogs varies with the location of skin sampled, method used to detect the organism, and disease status of the patient. In the only other study<sup>3</sup> that assessed the frequency of *Malassezia* spp detection on the periocular skin of dogs, the organism was identified by fungal culture of swab specimens of the periocular area in 3 of 33

(9%) healthy dogs and 24 of 54 (44%) dogs with pruritic cutaneous lesions at various anatomic locations; however, it is unclear how many of those dogs with pruritic lesions were affected in the periocular region. In the present study, cytologic evaluation identified *Malassezia* spp in 4 of 28 (14%) periocular samples obtained from eyes with blepharitis and 15 of 139 (11%) samples obtained from eyes without blepharitis. Because *Malassezia* spp were identified by fungal culture in that other study<sup>3</sup> and cytologic evaluation in the present study, the results of the 2 studies cannot be directly compared. Despite the fact that fungal culture is a more specific and sensitive method for identification of *Malassezia* spp than is cytologic evaluation,<sup>3</sup> we chose to use cytologic evaluation in the present study because it is used routinely in clinical practice and is easier to perform and less expensive than is a fungal culture.

In the present study, the proportion of periocular samples that contained *Malassezia* spp was slightly higher for eyes with blepharitis (4/28 [14%]), compared with that for eyes without blepharitis (15/139 [11%]); however, there was no significant association between blepharitis and the presence of *Malassezia* spp on the periocular skin and the majority (15/19) of samples in which *Malassezia* spp were identified were obtained from eyes without blepharitis. This was an unexpected finding because researchers have postulated that some microorganisms that colonize the periocular skin may cause blepharitis,<sup>9</sup> and the presence of *Malassezia* spp has been associated with blepharitis in humans<sup>10,11</sup> and dermatitis at sites other than the periocular region in dogs.<sup>5</sup>

To our knowledge, the present study is the first to evaluate the respective associations between the presence of *Malassezia* spp on the periocular skin of dogs and the presence and type of ocular discharge or the application and type of topical ophthalmic medications. Although the presence of *Malassezia* spp on the periocular skin of dogs was associated with the presence of mucoid or mucopurulent ocular discharge and the application of topical medications, particularly those that were aqueous-based, the design of the study was such that none of those associations could be defined as causative. Nevertheless, some hypotheses regarding potential mechanistic pathways for those associations can be made. For example, the positive association between the presence of *Malassezia* spp and mucoid or mucopurulent ocular discharge might indicate that the organism causes the ocular discharge or that the ocular discharge provides a favorable microenvironment for yeast colonization. Also, blepharitis could cause localized irritation that results in ocular discharge and have an indirect role in the association between the presence of *Malassezia* spp and ocular discharge. The topical administration of aqueous-based medications not considered immunosuppressive or the combination of oil- and aqueous-based medications that contained at least 1 potentially immunosuppressive drug most likely caused periocular overgrowth of *Malassezia* spp rather than vice versa, because none of the study dogs were administered topical ophthalmic medications for treatment of blepharitis or prior detection of *Malassezia* spp on the periocular skin. Instead, most study dogs had ocular conditions that necessitated treatment. Unlike the presence of *Malassezia* spp on the periocular skin, blepharitis was not significantly associated with the application of any type of medication assessed, and only 1 study dog devel-

oped blepharitis after topical treatment was initiated. These findings suggested that topical administration of some ophthalmic medications may be a risk factor for overgrowth of *Malassezia* spp on periocular skin but not for the development of blepharitis.

Interestingly, topical administration of oil-based or potentially immunosuppressive medications alone was not associated with the presence of *Malassezia* organisms in periocular samples or blepharitis in the present study. These findings might be the consequence of the small number of periocular samples in which *Malassezia* organisms were obtained from eyes that had blepharitis (n = 9) or were treated with an oil-based (9) or potentially immunosuppressive medication (9). Evaluation of a larger number of periocular samples that contained *Malassezia* spp would allow for a more detailed analysis of the respective associations between the presence of *Malassezia* spp and administration of specific potentially immunosuppressive drugs or ophthalmic medications compounded with commonly used oily vehicles such as corn, olive, coconut, or mineral oil. However, elucidation of a direct causal relationship between topical application of ophthalmic medications and the presence of *Malassezia* spp on the periocular skin will be difficult regardless of the size of the study population because any association between a specific topically administered medication and the presence of *Malassezia* spp on the periocular skin might be the consequence of a favorable environment for the yeast created by inflammation from allergic dermatitis caused by the active ingredient of the medication or its constituent compounds.<sup>12</sup> In human patients, allergic reactions to inactive constituents of ophthalmic medications are relatively common and have prompted the use of preservative- or carrier-free medications.<sup>13</sup>

Results of the present study also suggested that, although there was no association between the sex of the dog sampled and the identification of *Malassezia* spp on the periocular skin or development of ocular discharge, female dogs were more likely than were male dogs to develop blepharitis. To our knowledge, this is novel information for dogs and is contrary to findings for canine atopic dermatitis, for which a sex predisposition has not been identified.<sup>14</sup> These contradictory findings may reflect a difference in the neutered status of the 2 study populations. In the present study, the low number of sexually intact dogs prevented the effect of neutered status from being evaluated. Similarly, the effect of breed on the presence of *Malassezia* spp on the periocular skin or blepharitis could not be assessed because the dogs of the present study represented a diverse range of breeds, with each breed represented by only a small number of dogs. It is possible that assessment of a larger population might identify a breed predisposition for the presence of *Malassezia* spp on the periocular skin and blepharitis because there are breed predispositions for the development of canine atopic dermatitis<sup>14</sup> and blepharitis is associated with that disease.<sup>15</sup> The role of atopic dermatitis in the development of blepharitis was not evaluated in the present study. Dogs with atopic dermatitis are predisposed to development of secondary bacterial and fungal skin infections<sup>5</sup>; therefore, it is possible that the dogs in the present study that had *Malassezia* spp identified in periocular samples obtained from eyes with blepharitis, particularly those without a history

of administration of topical ophthalmic medications or concurrent ophthalmic disease, had atopic dermatitis.

Results of the present study suggested that cytologic evaluation of skin surface samples obtained by adhesive tape is a practical method for identification of *Malassezia* spp on the periocular skin of dogs. *Malassezia* spp were identified on the periocular skin of 5% (3/56) of dogs that had apparently normal skin with no ocular discharge and were not being treated with any topical ophthalmic medications. Also, *Malassezia* spp were more likely to be identified in periocular samples obtained from eyes that were being treated topically with aqueous-based ophthalmic medications that did not contain potentially immunosuppressive drugs or a combination of oil- and aqueous-based medications that contained at least 1 potentially immunosuppressive drug, compared with samples obtained from eyes that were not treated with topical medications or were treated only with oil-based medications. Thus, the periocular skin of dogs such as these that develop blepharitis during treatment with topical ophthalmic medications should be assessed for the presence of *Malassezia* spp, and this assessment can be done by cytologic evaluation.

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- a. Diff Quik Solution II, Polysciences Inc, Warrington, Pa.  
 b. Stata IC version 12, StataCorp LP, College Station, Tex.
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## References

- Guillot J, Bond R. *Malassezia pachydermatis*: a review. *Med Mycol* 1999;37:295–306.
- Kumar A, Singh K, Sharma A. Prevalence of *Malassezia pachydermatis* and other organisms in healthy and infected dog ears. *Israel J Vet Med* 2002;57:145–148.
- Cafarchia C, Gallo S, Romito D, et al. Frequency, body distribution, and population size of *Malassezia* species in healthy dogs and in dogs with localized cutaneous lesions. *J Vet Diagn Invest* 2005;17:316–322.
- Outerbridge CA. Mycologic disorders of the skin. *Clin Tech Small Anim Pract* 2006;21:128–134.
- DeBoer DJ, Marsella R. The ACVD task force on canine atopic dermatitis (XII): the relationship of cutaneous infections to the pathogenesis and clinical course of canine atopic dermatitis. *Vet Immunol Immunopathol* 2001;81:239–249.
- Rosales MS, Marsella R, Kunkle G, et al. Comparison of the clinical efficacy of oral terbinafine and ketoconazole combined with cephalexin in the treatment of *Malassezia* dermatitis in dogs—a pilot study. *Vet Dermatol* 2005;16:171–176.
- Bensignor E, Janikowski F, Seewald W, et al. Comparison of two sampling techniques to assess quantity and distribution of *Malassezia* yeasts on the skin of Basset Hounds. *Vet Dermatol* 2002;13:237–241.
- Williams RL. A note on robust variance estimation for cluster-correlated data. *Biometrics* 2000;56:645–646.
- Lee ST, Oh DH, Jung JY, et al. Comparative ocular microbial communities in humans with and without blepharitis. *Invest Ophthalmol Vis Sci* 2012;53:5585–5593.
- Derbel M, Benzina Z, Ghorbel I, et al. *Malassezia* fungal blepharitis: a case report. *J Fr Ophthalmol* 2005;28:862–865.
- Ninomiya J, Nakabayashi A, Higuchi R, et al. A case of seboreic blepharitis: treatment with itraconazole. *Jpn J Med Mycol* 2002;43:189–191.
- Pena MT, Leiva M. Canine conjunctivitis and blepharitis. *Vet Clin North Am Small Anim Pract* 2008;38:233–249.
- Bielory L. Ocular allergy guidelines: a practical treatment algorithm. *Drugs* 2002;62:1611–1634.
- Olivry T, DeBoer DJ, Favrot C, et al. Treatment of canine atopic dermatitis: 2010 clinical practice guidelines from the International Task Force on Canine Atopic Dermatitis. *Vet Dermatol* 2010;21:233–248.
- Griffin CE, DeBoer DJ. The ACVD task force on canine atopic dermatitis (XIV): clinical manifestations of canine atopic dermatitis. *Vet Immunol Immunopathol* 2001;81:255–269.