EVALUATION OF MICROFLORA ASSOCIATED WITH CANINE OTITIS EXTERNA

Roxana Topală, I. Burtan, M. Fîntînariu, S. Ciobanu, L.C. Burtan, Ioana Burcoveanu, E.V. Şindilar

Faculty of Veterinary Medicine Iasi, Romania,roxanatiron22@yahoo.com

Abstract

This study was undertaken to characterize otic microflora encountered in dogs with clinical signs of otitis externa and to determine its role in causing the disease. For this purpose 73 otic samples from normal dogs and 149 otic samples from dogs with different clinical stages of otitis were microbiological evaluated. The most common pathogens in the etiology of otitis include members of Staphylococcus genus, Streptococcus genus and yeast from Malassezia genus. From normal dogs Malassezia canis was isolated as a pure culture or with staphylococci and streptococci in 32 samples, representing 43.8%, staphylococci were recorded at a frequency of isolation (in pure and mixed cultures) of 32.9% and streptococci were isolated in 17 pure or mixed cultures, representing 23.3%. Staphylococcus aureus, Pseudomonas aeruginosa and Proteus spp. were not isolated in samples taken from dogs without ear problems. From dogs with varied clinical stages of otitis, Malassezia canis reported a frequency of isolation (in pure or mixed cultures) of 33.3%, staphylococci were isolated in 22.4% and streptococci in 19% from samples, in pure or mixed cultures. Staphylococcus aureus and Pseudomonas aeruginosa were isolated both in pure cultures and mixed cultures and Proteus spp. only in mixed cultures.

Key words: *canine, microflora, otitis, staphilococci, streptococci.*

INTRODUCTION

Canine otitis externa is one of the most common diseases encountered in veterinary practice and is estimated to affect between 5% and 20% of dogs (Gotthelf, 2005). Infectious otitis externa occurs as a secondary complication of primary factors that initiate inflammation within the external ear canal, such as hypersensitivity disorders (atopic dermatitis, food reactions, contact dermatitis), foreign bodies, ectoparasites, keratinization disorders, endocrine and autoimmune diseases (1,2,6).

Common pathogenic bacterial species include *Staphylococcus, Streptococcus, Pseudomonas, Proteus, Corynebacterium* and *Enterococcus* (2,4,5). The most common fungal pathogen in the etiology of otitis is

Malassezia spp., and rarely *Candida* or other saprophytic fungal organisms (7,8).
Considering multifactorial pathogenesis of otitis, treatments are varied and include topical therapy with antibiotic, antifungal or corticosteroid medication used alone or in combination (1,3,6). In canine otitis, the clinical diagnosis has an informative value, the adequate selection of a therapeutic method is conditioned by the results of the paraclinical diagnosis tests, imposed by the etiological polymorphism, especially microbiological examinations (2,8).
The purpose of the present study was to evaluate otic microflora diversity and involvement in emergence of various clinical forms of otitis.

MATERIALS AND METHODS

Cases were selected for study between January 2012 and October 2012 from dogs presented at Clinics of Faculty of Veterinary Medicine of Iasi, for ambulatory treatment, or hospitalization. Dogs of any age, breed or sex were eligible for enrollment.
The 73 healthy, normal dogs were selected as the control group, if there was no previous history of ear disease and no history of underlying disease (hypersensitivity disorders, keratinization disorders, endocrine and autoimmune diseases) and no clinical signs of ear or skin disease. Dogs in the control group were not currently on any medication other than preventive antiparasitic medication; dogs were not included if ototopical cleansers were used in the previous 2 weeks or systemic antibiotic/antifungal medication was administrated in the previous 4 weeks.
From dogs with different clinical stages of otitis, 149 samples were collected. Dogs were recruited based on history and clinical examination, which revealed the presence of characteristic signs (head shaking, local pain, pruritus, erythema, otorrhoea) in at least one ear. Complete physical and dermatological examinations were performed prior to collection of otic samples and physical examination findings were categorized to establish clinical stage of otitis. Dogs were not included if any topical or systemic therapy was administrated.
Samples were collected with a sterile culture swab introduced into ear canal and were sent to a private Microbiology Laboratory where they were processed according to conventional methods of isolation and identification.

RESULTS

From normal dogs (table 1) *Malassezia canis* was isolated as a pure culture or with staphylococci and streptococci in 32 samples, representing 43.8%, staphylococci were recorded at a frequency of isolation (in pure and mixed cultures) of 32.9% and streptococci were isolated in 17 pure or mixed cultures, representing 23.3%. *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Proteus* spp. were not isolated in samples taken from dogs without ear problems.

Table 1. Microorganisms isolation frequency from normal ear canal at dogs

Microorganism isolated	**No.**	**Frequency (%)**
Malassezia canis in pure culture	13	17.8
+*Staphylococcus* spp.	11	15
+*Streptococcus* spp.	8	11
+*Staphylococcus aureus*	-	-
+*Pseudomonas aeruginosa*	-	-
+*Proteus* spp.	-	-
Total	32	43.8
Staphylococcus spp. in pure culture	6	11
+*Streptococcus* spp.	5	6.9
+*Staphylococcus aureus*	-	-
+*Pseudomonas aeruginosa*	-	-
+*Proteus* spp.	-	-
Total	24	32.9
Streptococcus spp. in pure culture	4	5.4
+*Staphylococcus aureus*	-	-
+*Pseudomonas aeruginosa*	-	-
+*Proteus* spp.	-	-
Total	17	23.3
Staphylococcus aureus	-	-
Pseudomonas aeruginosa	-	-
Proteus spp.	-	-
TOTAL	**73**	**100**

Data in Table 2 reveals that *Malassezia canis* was isolated in 50 samples from dogs with varied clinical stages of otitis, representing 33.3%, of which 18 samples in pure culture. In mixed culture *Malassezia canis* was identified with staphylococci in 11 samples (7.4%), with streptococci in 9 samples (6%) and with *Staphylococcus aureus* in 8 samples (5.3%). *Malassezia*

canis recorded a frequency of isolation of 1.3%, both with *Pseudomonas aeruginosa* and with *Proteus* spp., respectively in 2 samples.
Staphylococci were isolated in 34 samples, representing 22.6%, from which 10% in pure culture. In mixed cultures *Staphylococcus* spp. recorded a isolation frequency of 6% with Streptococcus spp., 3.3% with *Staphylococcus aureus*, 2% with *Pseudomonas aeruginosa* and 1.3% with *Proteus* spp.

Table 2. Microorganisms isolation frequency from dogs with varied stages of otitis

Microorganism isolated	**Clinical stage**	**No.**	**Frequency (%)**
Malassezia canis in pure culture	C	18	12
+*Staphylococcus* spp.	E	11	7.4
+*Streptococcus* spp.	E	9	6
+*Staphylococcus aureus*	S	8	5.3
+*Pseudomonas aeruginosa*	S/U	2	1.3
+*Proteus* spp.	S/U	2	1.3
Total		50	33.3%
Staphylococcus spp. in pure culture	E	15	10
+*Streptococcus* spp.	E	9	6
+*Staphylococcus aureus*	S	5	3.3
+*Pseudomonas aeruginosa*	S/U	3	2
+*Proteus* spp.	S/U	2	1.3
Total		34	22.6%
Streptococcus spp. in pure culture	E	15	10
+*Staphylococcus aureus*	S	10	6.7
+*Pseudomonas aeruginosa*	S/U	3	2
Total		28	18.7%
Staphylococcus aureus in pure culture	S	2	1.3
Total		25	16.7%
Pseudomonas aeruginosa in pure culture	S	1	0.7
Total		9	6%
Proteus spp. in pure culture	S	-	-
Total		4	2.7%
TOTAL		**149**	**100%**

C – ceruminous otitis
E – exudative otitis
S – supurative otitis
U – ulcerative otitis

In pure and mixed cultures, *Streptococcus* spp. recorded a isolation frequency of 18,7%. In pure culture were isolated in 15 samples (10%), with *Staphylococcus aureus* in 10 samples (6.7%) and with *Pseudomonas aeruginosa* in 3 samples (2%).
In pure culture *Staphylococcus aureus* and *Pseudomonas aeruginosa* were isolated in 2, respectively in 1 sample, recording a total isolation frequency (in pure and mixed cultures) of 16.7%- *Staphylococcus aureus* and 6%- *Pseudomonas aeruginosa*.
Proteus spp. was identified only in mixed cultures in 4 samples (2 with *Malassezia canis* and 2 with *Staphylococcus* spp.), representing 2.7%.
Data obtained reveals that in ear canal of dogs there are commensal and pathogenic conditioned bacteria so that, the isolated and identified bacteria in otic samples collected from dogs with various forms of otitis did not confirm the determinative role in external otitis emergence, but only its favoring role. Microorganisms that are normal resident flora or opportunistic pathogen rapidly colonize the ear canal when microclimate is alterated in early clinical forms resulting progressive deterioration.

CONCLUSIONS

The most common pathogens in the etiology of otitis include members of *Staphylococcus* genus, *Streptococcus* genus and yeast from *Malassezia* genus.
Malassezia canis was isolated (in pure or mixed cultures), in samples collected from normal dogs and also from dogs with varied clinical stages of otitis.
Staphylococcus aureus, Pseudomonas aeruginosa and *Proteus* spp. were not isolated in samples taken from dogs without ear problems.
From dogs with otitis *Staphylococcus aureus* and *Pseudomonas aeruginosa* were isolated both in pure cultures and mixed cultures and *Proteus* spp. only in mixed cultures.
Comparative evaluation of ear flora (in health or disease) did not confirm the determinative role in external otitis emergence, but only its favoring role.

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