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**METHICILLIN-RESISTANT STAPHYLOCOCCI FROM DOGS AND CATS WITH DERMATOLOGIC PROBLEMS**

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**ABSTRACT**

Methicillin-resistant staphylococci, which are commonly found worldwide, known as one of the most important hospital infections and it is a problem in our country. The aim of the study was to examine the distribution of methicillin-resistant staphylococci (MRS) from dogs and cats with dermatologic problems. Pre-moistened swab samples were collected from skin lesions of 69 dogs and 23 cats, which were brought to the Microbiology Department for mycological examination during one year. Swabs were cultured onto Mannitol Salt Agar (supplemented with 5 µg/ml oxacillin) plates and incubated at 37° C for 24 hours. After incubation, identification of the methicillin-resistant isolates was performed by conventional methods and API Staph-Ident system. Methicillin resistance was detected phenotypically by the disc diffusion method and following genotypically by PCR for the detection of the *mecA* gene. Antibiotic susceptibility test was made according to CLSI standard by using penicillin G, amoxicillin/clavulanic acid, erythromycin, chloramphenicol, cephalothin, azithromycin, trimethoprim /sulphamethoxazole. 4.35 % of dogs with skin problems and 4.35 % of cats with skin problems are MRS positive. 66.7% of MRS positive dogs have methicillin-resistant *S. pseudintermedius*. Using empirical antimicrobials should be avoided, and veterinarians should consider MRS not only for the patient but also for epidemiological implications.

**Keywords:** Methicillin-resistant, dog, cat, staphylococci

**1. INTRODUCTION**

One of the most commonly recovered agents from pet medicine in veterinary clinical practice is Staphylococci. In addition to their presence on the skin and mucosal surfaces, they caused various diseases in animals [16]. Dogs and cats have become a part of modern society with the development of the world. Therefore the care and welfare of these animals gain importance.

Studies have shown that dogs are more susceptible to bacterial skin infections and have more cases than other domestic animals or humans [3], [18], [27]. The primary skin pathogens of dogs are *Staphylococcus* species of which *S. intermedius* (reclassified as *S. pseudintermedius*) is far the most important. Ninety per cent of canine pyoderma is related to staphylococci including *S. pseudintermedius*, *S. schleiferi*, and *S. aureus*. Coagulase-negative *Staphylococcus* spp. have also been associated with infections in people and dogs [10], [11], [15], [31].

Staphylococci do not generally cause a specific disease in cats, but many researchers have reported the cases of superficial pyoderma, bacterial folliculitis and superficial dermatitis caused by staphylococci in cats [8], [9], [13], [22]. *S. intermedius* is considered as a zoonotic pathogen and transmission between humans, and their pets can occur in both directions as well as *S. aureus* [3], [27], [29].

One of the most problematic resistant organism of modern times is methicillin-resistant staphylococci. All methicillin-resistant staphylococci (MRS) are, considered resistant to all  $\beta$ -lactam antibiotics [5], [18]. Therefore, the determination of methicillin resistance is very important for canine, owners and clinicians. Transfer of MRS from pet animals to man has also been documented. Consequently, MRS isolated from cats and dogs has also emerged importance for public health [22], [24]. The study aimed to examine the distribution of MR staphylococci from dogs and cats with dermatologic problems.

## **2. MATERIALS - METHODS**

### **Samples**

Pre-moistened swab samples were collected from skin lesions of 69 dogs and 23 cats, which were brought to the Microbiology Department for mycological examination during one year. Clinical signs and history of antimicrobial therapy of the sampled animals were noted.

### **Culture**

Swabs were cultured onto Mannitol Salt Agar (supplemented with 5  $\mu$ g/ml oxacillin) plates and incubated at 37° C for 24 hours. After incubation the colonies on MSA plates were examined macroscopically and microscopically, each colony with different macroscopic morphology were passaged onto Nutrient agar containing 7% blood to have pure cultures. Identification of the methicillin-resistant isolates was performed by conventional methods and API Staph-Ident system (API System, BioMérieux).

Differentiation of *S. pseudintermedius* from other members of *S. intermedius* group (SIG) has been acquired by PCR-restriction fragment length polymorphism (RFLP) tests by *Mbo*I enzyme [2].

### **Antibiotic Susceptibility Tests**

MR was detected phenotypically by the disc diffusion method [6] and following genotypically by PCR for the detection of the *mecA* gene [20]. Antibiotic susceptibility tests were performed according to the Clinical and Laboratory Standards Institute (CLSI) standard by using penicillin G (10 units), amoxicillin/clavulanic acid (20 $\mu$ g/ 10  $\mu$ g), erythromycin (15  $\mu$ g), chloramphenicol (30 $\mu$ g), cephalothin (30 $\mu$ g), azithromycin (15  $\mu$ g), trimethoprim /sulphamethoxazole (1.25  $\mu$ g/ 23.75  $\mu$ g) antibiotic discs which mainly used in the therapy of skin infections caused by *Staphylococcus* spp.

## **3. RESULTS**

While 14 of the dogs were under 1 year old, the remaining 55 dogs were between 1 and 12 years of age. Pathogen dermatophyte agents were recorded by mycological examination in 6 of the dogs. It was recorded that 30 of the dogs received antibiotic therapy for a certain period in the last year.

As a result of the bacteriological examination, 3 MRS strains were isolated out of 69 dogs. Two of the isolates were identified as *S. pseudintermedius* while one was *S. epidermidis*. *Microsporum canis* was also isolated from a dog in which *S. epidermidis* was isolated. No dermatophyte agent was isolated from the other two animals. One dog in which *S. pseudintermedius* was isolated showed only erythema, and the other one showed erythema, exfoliation and crusting. The cause of these signs might be the toxin (exfoliative) of the bacteria. All the MRS positive dogs have histories of antimicrobial therapy within one year of the period. While two of the cats were under one year old, the remaining 21 cats were between 1 and 15 years of age. Pathogen dermatophyte agents were determined by mycological examination in three of the cats.

As a result of the bacteriological examination, one MRS strain was isolated out of 23 cats. The feline isolate was identified as *S. arlettae*. Dermatophyte agent was not isolated, and there is no information about antimicrobial therapy of the cat. Antibiotic susceptibility tests results of canine and feline isolates are shown in table 1 and 2, respectively.

**Table1. Antimicrobial susceptibilities of the canine isolates**

Canine isolates	P 10	AMC 30	C 30	CO 25	AT 15	E 15	CH 30
<i>S. pseudintermedius</i>	R	R	I	R	R	R	R
<i>S. pseudintermedius</i>	R	R	S	R	R	R	R
<i>S. epidermidis</i>	R	S	R	R	R	R	S

P10= Penicillin G (10 µg); AMC 30 10= Amoxicillin/Clavulanic acid (20/10 µg); C 30= Chloramphenicol (30µg); CO 25= Trimethoprim /Sulphamethoxazole (25 µg/ml); AT 15= Azithromycin (15 µg/ml); E15= Erythromycin (15 µg); CH 30= Cephalothin (30 µg); I: intermediate, S: susceptible, R: resistant

**Table 2. Antimicrobial susceptibility of the feline isolate**

Feline isolate	P 10	AMC 30	C 30	CO 25	AT 15	E 15	CH 30
<i>S. arlettae</i>	R	R	S	R	R	R	R

P10= Penicillin G (10 µg); AMC 30 10= Amoxicillin/Clavulanic acid (20/10 µg); C 30= Chloramphenicol (30µg); CO 25= Trimethoprim /Sulphamethoxazole (25 µg/ml); AT 15=

Azithromycin (15 µg/ml); E15= Erythromycin (15 µg); CH 30= Cephalothin (30 µg); R: resistant, S: susceptible

#### **4. DISCUSSION**

Methicillin-resistant staphylococci, which are commonly found worldwide, known as one of the most important hospital infections and it is a problem in our country. *Staphylococcus pseudintermedius* are considered the most important pathogen in dogs' skin infections. Consequently, in the whole world, the incidence of methicillin-resistant *Staphylococcus pseudintermedius* (MRSP) is increasing and the isolation of agents from the skin of dog owners and veterinarians increases the fear of treatment [1]. In the previous studies, the prevalence of MRSP in dogs' skin varied notably between countries and ranged from 0% to 7% [12]. However, in recent studies, the increased prevalence of MRSP in dogs and cats (0-66 %) was highlighted, in addition, dogs and cats have been considered as potential reservoirs that seriously threaten animal and public health by limiting the available antibiotic selection [4], [14], [17], [26], [28], [30]. In this study, two of the canine isolates were identified as *S. pseudintermedius*. They were found to be low with 4.35 % prevalence compared to other studies.

As indicated in many studies, opportunistic coagulase-negative staphylococci frequently have multiple antimicrobial resistances [4], [8], [9], [15], [19], [28]. Antibiotic use in patients with multi-resistance strain it may cause resistance to other antimicrobial groups. Tabatabaei et al. (2019) reported that multidrug-resistant (MDR) MRSA and MDR-MRSP isolated from concomitant animals had been reported to be highly resistant to tetracycline and erythromycin. Loeffler and Lloyd (2018) indicated that MRSP's emergence in dogs and even the presence of MDR is a greater challenge for veterinary medicine. As similar with other researches, MRSP isolates in this study were MDR. This may be a severe problem for veterinarians as they limit the options for antibiotic treatment.

In this study, methicillin-resistant *S. epidermidis* was detected from a dog in which also *M. canis* was isolated as consistent with the indicated literature. Mazel and Davies (1999) have reported that the access of staphylococci to a full gene pool on the skin and mucosal surfaces can favour the acquisition of resistance genes. Öztürk et al. (2010) have determined that methicillin-susceptible or resistant *S. aureus* and *S. intermedius* are the predominant organisms in dogs with otitis externa and skin infections in dogs. Moreover, they noticed the importance of correct diagnosis of MR for the treatment of dogs.

Magalhaes et al. (2010) has described risk factors for MRSA infection in dog and cats. The researchers identified the numerous risk factors including the number of antimicrobial courses, number of days admitted to the veterinary practices, ongoing infection, and surgical implant. Also, Fitzgerald (2009) has reported that canine atopic dermatitis alters the availability of cutaneous receptors for staphylococci and facilitates bacterial adherence. All the MRS positive dogs have histories of antimicrobial therapy, and mycotic infection was identified from one of the dogs while there is no information about the antimicrobial treatment of the cat.

#### **5. CONCLUSIONS**

4.35 % of dogs with skin problems and 4.35 % of cats with skin problems are MRS positive. 66.7% of MRS positive dogs were identified as methicillin-resistant *S. pseudintermedius*. This high rate indicates that the emergence and spread of these species will be a major problem for veterinary medicine and society. Using empirical antimicrobials should be avoided, and veterinarians should consider MRS not only for the patient but also for epidemiological implications.

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