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ScienceDirect
Veterinary Microbiology 126 (2008) 277–281
veterinary microbiology
www.elsevier.com/locate/vetmic

Short communication

Methicillin-resistant staphylococcal colonization in dogs entering a veterinary teaching hospital

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Received 28 March 2007; received in revised form 15 June 2007; accepted 18 June 2007

Abstract

Nasal, axillary and rectal swabs were collected from 193 dogs admitted to the Ontario Veterinary College Veterinary Teaching Hospital. Enrichment culture was performed and coagulase positive staphylococci were identified via standard methods. Methicillin-resistant *Staphylococcus pseudintermedius* was isolated from 4/193 (2.1%) dogs, and methicillin-resistant *Staphylococcus aureus* and methicillin-resistant *Staphylococcus schleiferi* subsp. *coagulans* were each isolated from 1/193 (0.5%) dogs. Methicillin-resistant *Staphylococcus intermedius* was not identified. All *S. pseudintermedius* isolates were unrelated on pulsed-field gel electrophoresis. Evaluation of the epidemiology of methicillin-resistant staphylococcal colonization is necessary to understand the apparent emergence of these strains and to develop appropriate control strategies.

Keywords: *Staphylococcus pseudintermedius*; *Staphylococcus intermedius*; *Staphylococcus aureus*; Methicillin-resistance; *Staphylococcus schleiferi*

Coagulase positive staphylococci, particularly *Staphylococcus aureus*, *Staphylococcus intermedius* and *Staphylococcus schleiferi* subsp. *coagulans*, are opportunistic pathogens that are of concern in veterinary medicine. Methicillin-resistant strains of these organisms are being reported with increasing frequency in dogs (Baptiste et al., 2005; Jones et al., 2007; Loeffler et al., 2005; Morris et al., 2006; Weese et al., 2006), which has caused concern both for treatment of animal disease and potential public health consequences. Methicillin-resistant *S. aureus* (MRSA) is of particular concern and there have been increasing reports of infections in dogs and cats both in veterinary hospitals and in the community (Baptiste et al., 2005; Loeffler et al., 2005; Morris et al., 2006; Tomlin et al., 1999; Weese et al., 2006). Suspected transmission of MRSA between people and pets has been reported (Leonard et al., 2006; Loeffler et al., 2005; van Duijkeren et al., 2004; Weese et al., 2006). Methicillin-resistant strains of *S. intermedius* (MRSI) and *schleiferi* subsp. *coagulans* (MRSS) have also been identified in dogs, particularly in skin and soft tissue infections (Jones et al., 2007; Morris et al.,

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doi:10.1016/j.vetmic.2007.06.015

2006). Recently, methicillin-resistant *Staphylococcus pseudintermedius* (MRSP) infections were reported in dogs (Devriese et al., 2005; Sasaki et al., 2007). The risk of zoonotic transmission of these species is unclear, however, transmission of *S. intermedius* between dogs and their owners has been reported (Guardabassi et al., 2004).

The purpose of this study was to determine the prevalence of methicillin-resistant coagulase positive staphylococci colonization in dogs upon admission to a veterinary teaching hospital and to identify risk factors associated with colonization.

A convenience sample of 193 dogs was studied at the Ontario Veterinary College Veterinary Teaching Hospital (OVC-VTH) over a 2-month period (October and November) in 2004. All ages, genders and breeds of dogs were eligible for inclusion. Owner consent was obtained prior to enrolment of each dog.

At the time of admission, cotton swabs moistened with sterile saline were used to collect nasal, axillary and rectal samples from each dog. Nasal swabs were collected by inserting the swab 0.5–1 cm into the nostrils, or by rubbing the swab over the external nares in the case of animals where the nares were too small for swab insertion. Rectal swabs were collected by inserting swabs 1 cm into the rectum. Axillary swabs were collected by rubbing the swab in the axilla. The swabs were placed in liquid Stuart's medium and maintained at 4 °C until processing. Owners completed a brief questionnaire about their dog that included gender, age, breed, presenting complaint as well as history of previous surgery, antimicrobial use, hospitalization or immunosuppressive therapy within the past 3 months.

The swabs were plated directly onto mannitol-salt agar with 2 μg/mL oxacillin and incubated at 35 °C for 48 h. In addition, swabs were placed in 2 mL of enrichment broth consisting of 10 g/L Tryptone T, 75 g/L sodium chloride, 10 g/L mannitol and 2.5 g/L yeast extract, for 24 h at 35 °C. An aliquot of 100 μL broth was inoculated onto mannitol-salt agar with 2 μg/mL oxacillin as above. Isolates were identified as coagulase positive staphylococci based on colony morphology, Gram stain appearance, positive catalase reaction and positive tube coagulase test. *S. aureus* was identified by latex agglutination test (LAT) (Pastorex Staph Plus, Bio-Rad Laboratories Ltd., Mississauga, Canada). Presumptive identification of *S. intermedius* and *S. schleiferi* were performed by biochemical methods, consisting of maltose and trehalose fermentation, and polymyxin B susceptibility testing. Confirmation of identification was performed by sequencing of the 16s rRNA gene. Methicillin-resistance was confirmed by demonstration of penicillin binding protein (PBP) 2a using a latex agglutination test (Denka Seinken Co. Ltd., Tokyo, Japan) and growth on Mueller Hinton agar with 6 μg/mL oxacillin. MRSA was typed via *Sma*I PFGE and categorized as Canadian epidemic MRSA (CMRSA) types as previously described (Mulvey et al., 2001). All methicillin-resistant isolates were tested for Panton-Valentine leukocidin (PVL) genes using real time PCR (Rankin et al., 2005).

Follow-up investigation was performed 1 month following hospital discharge on an Irish wolfhound that was identified as colonized with methicillin-resistant *S. pseudintermedius*. All dogs in the kennel, the dog owners, kennel environment, referral veterinary clinic staff and clinic environment were sampled. Nasal and rectal swabs were collected from each dog, nasal swabs were collected from each person and electrostatic wipes (Swiffer™) were used to sample the kennel and clinic environments. Isolation of methicillin-resistant, coagulase positive staphylococci was performed as described above, with the exception that electrostatic cloths were enriched using 50 mL of enrichment broth.

The prevalence of methicillin-resistant staphylococci colonization in dogs was calculated. Based on the number of colonized dogs, categorical comparisons were performed using Chi-square analysis or Fisher's exact test. Risk factors for colonization were evaluated via stepwise forward logistic regression. A P-value of <0.05 was considered significant for all comparisons.

This study was approved by the University of Guelph Research Ethics Board and Animal Care Committee.

Nasal, axillary and rectal swabs were collected at admission from 193 dogs. Dogs enrolled in the study presented to medicine ($n = 34$), oncology ($n = 33$), dermatology ($n = 34$), surgery ($n = 33$), neurology ($n = 21$), emergency ($n = 21$), cardiology ($n = 13$), theriogenology ($n = 3$) and dentistry ($n = 1$) services. Fifty-nine breeds were enrolled in the study with the Labrador retriever ($n = 20$), Golden retriever ($n = 19$),