

# Antimicrobial Susceptibility and rRNA Gene Restriction Patterns among *Staphylococcus intermedius* from Healthy Dogs and from Dogs Suffering from Pyoderma or Otitis Externa

By K. Pedersen<sup>1</sup> and H. C. Wegener<sup>2</sup>

<sup>1</sup>The Royal Veterinary and Agricultural University, Department of Veterinary Microbiology, Frederiksberg, and <sup>2</sup>Danish Veterinary Laboratory, Copenhagen, Denmark.

**Pedersen, K. and H. C. Wegener: Antimicrobial susceptibility and rRNA gene restriction patterns among *Staphylococcus intermedius* from healthy dogs and from dogs suffering from pyoderma or otitis externa. Acta vet. scand. 1995, 36, 335-342.** – A total of 60 *Staphylococcus intermedius* strains from dogs were investigated by their sensitivity to various antibiotics (50 strains) and by their rRNA gene restriction patterns (ribotyping) (60 strains). Fifteen isolates were from healthy dogs, 9 with otitis externa, and 36 with pyoderma, including 10 strains from a previous study. Sixty per cent of the 50 strains tested for antibiotic susceptibility demonstrated resistance to penicillin, 24% to spiramycin, 20% to tetracycline, 16% to chloramphenicol, and 2% to fucidic acid. All isolates were susceptible to amoxycillin with clavulanic acid, enrofloxacin, and sulphonamides with trimethoprim. There were no significant differences in antimicrobial susceptibility patterns observed among isolates from pyoderma, otitis externa or healthy dogs. Among the 60 strains studied by ribotyping, 10 different ribotypes were identified: 6 different ribotypes among isolates from otitis externa, 8 among isolates from pyoderma, and 5 among isolates from healthy dogs. One ribotype (profile C) was dominant among the isolates from healthy dogs while another ribotype (profile A) was dominant among strains from dogs suffering from pyoderma. This profile was not demonstrated in any of the strains from healthy dogs. From 5 different dogs suffering from pyoderma, 2 different clones were demonstrated based on their plasmid profile and antibiogram. In these dogs 1 of the clones always belonged to ribotype A. The results concerning strains of *S. intermedius* isolated from furunculosis suggest the existence of distinct subpopulations with different pathogenicity to dogs.

*antibiotic resistance; ribotyping.*

## Introduction

*Staphylococcus intermedius* was first described as a new species by Hájek in 1976. The name was given to designate that it possesses some biochemical and cell wall properties of *Staphylococcus aureus* and some of *Staphylococcus epidermidis*, and therefore holds an intermediate position between these 2 species.

Important characters to differentiate between the 3 species or to differentiate them from other staphylococcal species are outlined by Hájek (1976), Phillips & Kloos (1981) and Schleifer (1986).

Since then, *S. intermedius* has been isolated from several animal species, including dogs, cats, cows, goats, horses, monkeys, humans,

pigeons, rats, and mink (Adegoke 1986, Berg *et al.* 1984, Biberstein *et al.* 1984, Hajek 1976, Raus & Love 1983) from normal animals and as the cause of various pathological conditions. In dogs, *S. intermedius* is the most important pathogenic staphylococcal species, having been isolated from a variety of clinical manifestations, such as otitis externa, purulent dermatitis, wound infections, abscesses, osteomyelitis, metritis, mastitis, respiratory tract infections, and bacteraemia (Biberstein *et al.* 1984, Raus & Love 1983) and it has been shown to be intimately associated with canine furunculosis or pyoderma (DeBoer 1990, Muller *et al.* 1989, Quadros 1974, Wegener & Pedersen 1992), a chronic skin disorder in dogs. Recently, it was found that *S. intermedius* was commonly present in the canine nasal cavity (Fukuda *et al.* 1984) and the gingival flora (Talan *et al.* 1989a) and although the bacterium was rarely isolated from healthy humans (Talan *et al.* 1989b) it was occasionally responsible for dog bite wound infections in humans (Talan *et al.* 1989a). Lee (1994) described that in 34 cases of dog-bite wounds in humans treated at Public Health Laboratory, Derriford Hospital, Plymouth, England, *S. intermedius* was found in 6 patients. The bacterium may therefore be considered a potential zoonotic pathogen (Talan *et al.* 1989a,b, Lee 1994). However, the bacterium has also been isolated from infections in humans without association with animals. Thus, Lee (1994) described 2 cases of infection with *S. intermedius* in elderly patients with clinically infected varicose leg ulcers, and in one 13 year old patient with infected suture lines. The author suggested that more human infections with *S. intermedius* were likely to occur, but were wrongly diagnosed as the closely related *S. aureus*. Like *S. aureus*, *S. intermedius* has been shown to produce enterotoxins, although in lower amounts (Fukuda *et*

*al.* 1984) and the first documented report on *S. intermedius* as the cause of food poisoning in more than 265 cases in western USA was recently presented (Khambaty *et al.* 1994). The authors demonstrated clonality among the isolated strains by pulsed-field gel electrophoresis.

Infections with *S. intermedius* are generally treated with antibiotics. It has been shown that susceptibility patterns of *S. intermedius* to various antimicrobials, in general are similar to those reported for *S. aureus* of various origin (Biberstein *et al.* 1984, Woldehiwet & Jones 1990). For the treatment of chronic or recurrent furunculosis, autochthonous vaccines are additionally frequently attempted.

In a previous study we investigated the clonal composition of *S. intermedius* isolated from pathological processes of dogs suffering from pyoderma (Wegener & Pedersen 1992). It was concluded that more than 1 clone could be present, an observation previously recognized for *Staphylococcus hyicus* isolated from exsudative epidermitis in pigs (Wegener 1992), and the impact of this finding in relation to antibacterial and immunostimulant therapy was discussed.

Previous work on canine pyoderma have focused mainly on host-related triggering factors. The purpose of the present investigation was to study characteristics associated with the infectious agent, *S. intermedius*, by comparing isolates from healthy dogs with those from otitis externa and pyoderma using antibiotic resistance phenotypes (antibiograms) and ribotyping.

## Materials and methods

### *Sampling procedure and culture conditions*

Samples from 30 healthy dogs were collected with sterile cotton swabs from the nasal cavity, the tonsils, and the skin, while samples from dogs suffering from otitis externa or pyo-

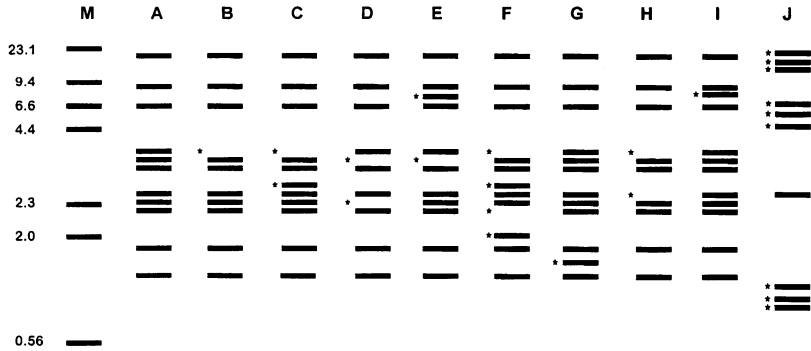


Figure 1. *Eco*RI rRNA gene restriction patterns (ribotypes) observed among 60 canine isolates of *Staphylococcus intermedius*, 36 isolates with pyoderma, 9 with otitis externa, and 15 isolates from healthy dogs. Different ribotypes were arbitrarily designated letters A through J. Lane M: DNA from phage lambda digested with *Hind*III served as molecular weight marker.

\* indicates band variations from index profile A.

derma were collected with sterile cotton swabs from typical lesions. Swabs were streaked onto blood agar (Columbia agar, Oxoid CM331, supplemented with 5% sterile bovine blood) and agar plates were incubated overnight at 37°C. Typical colonies were picked and bacteria were identified as *S. intermedius* according to Wegener & Pedersen (1992). Collected strains were stored in agar slants until used. A total of 60 *S. intermedius* strains were used for further investigation. Fifteen strains originated from the healthy dogs, while 9 and 26 strains, respectively, were isolated from dogs suffering from otitis externa or pyoderma. The remaining 10 strains, 2 from each of 5 dogs suffering from pyoderma, were isolated in a previous investigation (Wegener & Pedersen 1992). In these 5 patients, 2 different strains had been identified based on antibiogram and plasmid profile.

#### Antibiotic sensitivity testing

Strains were tested for their susceptibility to penicillin, tetracycline, fucidic acid, sulphona-

mides + trimethoprim, chloramphenicol, spiramycin, enrofloxacin, and amoxicillin + clavulanic acid by the disc diffusion method as previously described (Casals & Pringler 1991, Wegener & Pedersen 1992).

#### Ribotyping

Bacterial DNA was obtained by a method modified from Ausubel *et al.* (1989). Briefly, 1.5 ml of overnight cultures grown in tryptic soy broth (Difco) supplemented with 10 g yeast extract per l were centrifuged and pellet resuspended in 567 µl TE buffer (tris 10 mM, EDTA 1 mM, pH 8.0 added 125 U lysostaphin (Sigma). After incubation at 37°C for 1 h, 30 µl 10% SDS and 6 µl proteinase K, 10 mg/ml, were added and incubation was continued for another 1 h. Subsequently, impurities were precipitated by the addition of hexadecyltrimethylammonium bromide (cetyltrimethylammonium bromide, CTAB) (FLUKA Chemie, Switzerland), 10% w/vol in 0.7 M NaCl, and DNA was further purified by extraction with phenol:chloroform:isoamylalcohol (25:24:1) followed by chloroform:isoamylal-

Table 1. Antibiotic resistance of *Staphylococcus intermedius* from dogs suffering from pyoderma or otitis externa and from healthy dogs.

Antibiotic	Pyoderma, N = 26			Otitis externa, N = 9			Healthy dogs, N = 15			Total, N = 50		
	S <sup>a</sup> , n (%)	I <sup>a</sup> , n (%)	R <sup>a</sup> , n (%)	S, n (%)	I, n (%)	R, n (%)	S, n (%)	I, n (%)	R, n (%)	S, n (%)	I, n (%)	R, n (%)
Amoxicillin + clavulanic acid	26 (100)	0 (0)	0 (0)	9 (100)	0 (0)	0 (0)	15 (100)	0 (0)	0 (0)	50 (100)	0 (0)	0 (0)
Chloramphenicol	23 (88)	0 (0)	3 (12)	9 (100)	0 (0)	0 (0)	10 (67)	0 (0)	5 (33)	42 (84)	0 (0)	8 (16)
Enrofloxacin	26 (100)	0 (0)	0 (0)	9 (100)	0 (0)	0 (0)	15 (100)	0 (0)	0 (0)	50 (100)	0 (0)	0 (0)
Fucidic acid	25 (96)	0 (0)	1 (4)	9 (100)	0 (0)	0 (0)	15 (100)	0 (0)	0 (0)	49 (98)	0 (0)	1 (2)
Penicillin	11 (42)	0 (0)	15 (58)	4 (44)	0 (0)	5 (56)	5 (33)	0 (0)	10 (67)	20 (40)	0 (0)	30 (60)
Spiramycin	19 (73)	0 (0)	7 (27)	7 (78)	0 (0)	2 (22)	12 (80)	0 (0)	3 (20)	38 (76)	0 (0)	12 (24)
Tetracyclin	20 (77)	0 (0)	6 (23)	6 (67)	0 (0)	3 (33)	14 (93)	0 (0)	1 (7)	40 (80)	0 (0)	10 (20)
Sulphonamides + trimethoprim	26 (100)	0 (0)	0 (0)	9 (100)	0 (0)	0 (0)	15 (100)	0 (0)	0 (0)	50 (100)	0 (0)	0 (0)

<sup>a</sup>S = sensitive, I = intermediate, R = resistant, n = number in each sensitivity group.

cohol (24:1). Total bacterial DNA was precipitated by the addition of 0.6 volume isopropanol, washed once in 1 ml 70% ethanol and resuspended in 100 µl TE (10:1). DNA, 2–6 µg, was digested with 2 units of *EcoRI* (Amersham) at 37°C for 18 h, and subjected to electrophoresis in 0.8% agarose gels in TAE buffer. Following separation by electrophoresis, DNA fragments were blotted onto nylon hybridization membranes as described by *Southern* (1975). Membranes were hybridized with a digoxigenin labeled cDNA probe complementary to *Escherichia coli* 16S and 23S rRNA, prepared by reverse transcription according to the instructions of the manufacturer (Boehringer, Mannheim, Germany). Hybridized fragments were visualized by using a digoxigenin staining kit (Boehringer).

## Results

The results of the antibiotic sensitivity testing are shown in Table 1. All isolates were sensitive to amoxicillin + clavulanic acid, enrofloxacin, and sulphonamides + trimethoprim, while all strains, except 2, were sensitive to fucidic acid. Only 40% of the strains were sensitive to penicillin. No significant differences were recorded between strains from healthy dogs and dogs suffering from pyoderma or otitis externa.

A total of 10 different ribotypes were demonstrated among the 60 strains (Table 2). In contrast to the results of the antibiograms, an inhomogeneous distribution of the profiles was recognized within as well as among groups. In the 15 strains from healthy dogs, 5 different ribotypes were found, profile C being the most predominant while 6 different profiles were demonstrated among the 9 isolates from otitis externa. Among the 26 strains from pyoderma, 6 different profiles were recorded.

Table 2. Distribution of ribotypes of *Staphylococcus intermedius* among healthy dogs and dogs suffering from otitis externa or pyoderma.

Source	No.	Ribotype <sup>a</sup>									
		A	B	C	D	E	F	G	H	I	J <sup>b</sup>
Healthy dogs	15		2	8	1	3					1
Otitis externa	9	2	1	3			1	1			1
Pyoderma	26	11	1	6		4			1		3
Patient 1 <sup>c</sup>	2	1								1	
Patient 2	2	1	1								
Patient 3	2	1		1							
Patient 4	2	1			1						
Patient 5	2	1		1							

a) Ribotypes were arbitrarily designated letters A through J.

b) Profile J was very different from the other *S. intermedius* ribotypes.

c) Patients 1–5 were suffering from furunculosis, and from these patients, 2 different clones of *S. intermedius* based on their plasmid profile and antibiogram, were isolated (Wegener & Pedersen 1992).

Among these, profile A was the most frequently recognized (11 strains) followed by profile C (6), E (4), and J (3). Ribotype A was not present in any of the strains from healthy dogs.

In the 5 patients from which *S. intermedius* strains with 2 different antibiograms and plasmid profiles had been isolated, 1 of these strains always belonged to ribotype A.

The ten ribotypes are shown in Fig. 1. Profiles A through I displayed much the same pattern and differed from each other with only 1 or a few bands. In contrast, profile J was very deviating from all the other profiles. The significance of this observation is at present not known.

## Discussion

Several antibiotic susceptibility studies have been carried out for *S. intermedius*. Usually, resistance to penicillin and amoxicillin is very high, more than 50%, while resistance to tetracycline is often approximately 20–40%. Resistance to sulphonamides and trimethop-

rim may also be relatively high, >20%, while resistance to other antibiotics has usually been low (Biberstein *et al.* 1984, Cox *et al.* 1984, Greene & Schwarz 1992, Love 1989, Medleau *et al.* 1986, Woldehiwet & Jones 1990). In the present study we found 60% of the isolates resistant to penicillin while 20% were resistant to tetracycline, a result in accordance with previous reports. All strains, however, were sensitive to sulphonamides with trimethoprim. Greene & Schwarz (1992) found no differences in antibiotic resistance between clinical isolates and isolates from healthy dogs, a finding confirmed by the present results. Interestingly, Noble & Kent (1992) reported from a study of *S. intermedius* from cases of pyoderma in dogs that multiresistant strains occurred more commonly in previously treated cases than in primary consultations and more commonly in deep than superficial lesions suggesting a selection of resistant strains by antibiotic treatment.

The ecology and epidemiology of *S. intermedius* on the skin of dogs is at present not fully

understood. It has been shown that the bacterium is present on the skin, the nasal and oral mucosa, and the anal region on healthy dogs as well as on dogs suffering from pyoderma but counts on the diseased dogs are likely to be higher than those on healthy dogs (Allaker *et al.* 1992a,b, Berg *et al.* 1984, Devriese & DePelsmaecker 1987, Lloyd *et al.* 1991). The bacteria are not distributed equally: in some regions of the skin, counts tend to be higher than in others (Allaker *et al.* 1992b). In the present study, *S. intermedius* was demonstrated in healthy dogs more frequently in the nasal cavity and in the tonsils than on the skin, using a cotton swab technique (data not shown).

Healthy skin appears to be relatively resistant to infection with *S. intermedius* and the presence of *S. intermedius* on the skin of healthy dogs is probably transient caused by continuous contamination from the nose, the mouth, or the anal region and not the consequence of a colonization (Allaker *et al.* 1992a, Devriese & DePelsmaecker 1987). However, once changes in the microecology of the skin has occurred caused by conditions such as eczema, seborrhoea, or wounds, the injured skin constitutes a site, susceptible to colonization, proliferation, and subsequent infection (Allaker *et al.* 1992a,b, Lloyd *et al.* 1991). Much effort has hitherto been put into the study of factors which render the host more susceptible to infection. In contrast, we have chosen to investigate characters of the infectious agent to study if certain features could make some *S. intermedius* more likely to cause infection than others. In an earlier experiment we studied the clonality of *S. intermedius* isolated from dogs with pyoderma and concluded that more than 1 strain could be isolated from a typical lesion. In the present study we demonstrated notable differences in ribotype patterns between isolates from he-

althy dogs and those isolated from lesions of dogs suffering from pyoderma. The significance of this is at present not clear. Further investigations should clarify if certain subpopulations, distinguishable by their ribotypes, possess virulence factors related to canine pyoderma. Our results are in contrast to previous reports on the subject. Thus, in an earlier study Allaker *et al.* (1991) were unable to detect any differences between *S. intermedius* isolated from pyodermas and from healthy dogs with respect to a limited selection of virulence factors. Similarly, in a recent and important study, Hesselbarth *et al.* (1994) used pulsed field gel electrophoresis to study *S. intermedius* strains isolated from dogs: 28 strains from superficial pyoderma and 26 from healthy dogs (vaginal (9), nasal (8), and skin (9) isolates). The authors reported that all fingerprints were different although very homogeneous, but they were not able to detect any sub-populations of strains connected with infections and they concluded that such sub-populations probably don't exist. In our study, the strains from pyodermas were – in contrast to the study of Hesselbarth *et al.* (1994) – not isolated from superficial infections but from deeper infections (furunculosis) and it should be noted that in our study no connection between otitis externa and the ribotypes of the isolated *S. intermedius* strains could be demonstrated. The results may therefore suggest that the possible existence of sub-populations of *S. intermedius* only concerns certain kinds of infections. Further studies should be carried out to investigate this.

#### Acknowledgements

The technical assistance of Miss Sussi Kristoffersen and the financial support of "Fondet til sygdomsbekæmpelse hos vore familiedyr" is gratefully acknowledged.

## References

- Adegoke GO*: Characteristics of staphylococci isolated from man, poultry and some other animals. *J. appl. Bacteriol.* 1986, *60*, 97-102.
- Allaker RP, Lamport AI, Lloyd DH, Noble WC*: Production of "virulence factors" by *Staphylococcus intermedius* isolates from cases of canine pyoderma and healthy carriers. *Microb. Ecol. Health Dis.* 1991, *4*, 169-173.
- Allaker RP, Lloyd DH, Bailey RM*: Population sizes and frequencies of staphylococci at mucocutaneous regions on clinically normal dogs. *Vet. Rec.* 1992a, *130*, 303-304.
- Allaker RP, Lloyd DH, Simpson AI*: Occurrence of *Staphylococcus intermedius* on the hair and skin of normal dogs. *Res. Vet. Sci.* 1992b, *52*, 174-176.
- Ausubel FM, Brent R, Kingston RE*: Current protocols in molecular biology. Greene Publishing Associates and Wiley Interscience, New York, 1989.
- Berg JN, Wendell DE, Vogelweid C, Fales WH*: Identification of the major coagulase-positive *Staphylococcus* sp of dogs as *Staphylococcus intermedius*. *Amer. J. Vet. Res.* 1984, *45*, 1307-1309.
- Biberstein EL, Jang SS, Hirsh DV*: Species distribution of coagulase-positive staphylococci in animals. *J. clin. Microbiol.* 1984, *19*, 610-615.
- Casals JB, Pringler N*: Antibacterial sensitivity testing using Neo-Sensitabs. Rosco Diagnostica, Taastrup, Denmark, 1991.
- Cox HU, Hoskins JD, Roy AF, Newman SS*: Antimicrobial susceptibility of coagulase-positive staphylococci isolated from Louisiana dogs. *Amer. J. vet. Res.* 1984, *45*, 2039-2042.
- DeBoer DJ*: Strategies for management of recurrent pyoderma in dogs. In: *DeBoer DJ* (ed.): Advances in clinical dermatology. The Veterinary Clinics of North America: Small Animal Practice 1990, *20*, 1509-1524, WB Saunders Company, Philadelphia.
- Devriese LA, DePelsmaecker K*: The anal region as a main carrier site of *Staphylococcus intermedius* and *Streptococcus canis* in dogs. *Vet. Rec.* 1987, *121*, 302-303.
- Fukuda S, Tokuno H, Ogawa H, Sasaki M, Kishimoto T, Kawano J, Shimizu A, Kimura S*: Enterotoxigenicity of *Staphylococcus intermedius* strains isolated from dogs. *Zbl. Bakt. Hyg. A.* 1984, *258*, 360-367.
- Greene RT, Schwartz S*: Small antibiotic resistance plasmids in *Staphylococcus intermedius*. *Zbl. Bakt.* 1992, *276*, 380-389.
- Hájek V*: *Staphylococcus intermedius*, a new species isolated from animals. *Int. J. syst. Bacteriol.* 1976, *26*, 401-408.
- Hesselbarth J, Witte W, Cuny C, Rohde R, Amtsberg G*: Characterization of *Staphylococcus intermedius* from healthy dogs and cases of superficial pyoderma by DNA restriction endonuclease patterns. *Vet. Microbiol.* 1994, *41*, 259-266.
- Khambati FM, Bennett RW, Shah DB*: Application of pulsed-field gel electrophoresis to the epidemiological characterization of *Staphylococcus intermedius* implicated in a food-related outbreak. *Epidemiol. Infect.* 1994, *113*, 75-81.
- Lee J*: *Staphylococcus intermedius* isolated from dog-bite wounds. *J. Infect.* 1994, *29*, 105.
- Lloyd DH, Allaker RP, Pattinson A*: Carriage of *Staphylococcus intermedius* on the ventral abdomen of clinically normal dogs and those with pyoderma. *Vet. Dermatol.* 1991, *2*, 161-164.
- Love DN*: Antimicrobial susceptibility of staphylococci isolated from dogs. *Aust. vet. Practit.* 1989, *19*, 196-200.
- Medleau L, Long RE, Brown J, Miller WH*: Frequency and antimicrobial susceptibility of *Staphylococcus* species isolated from canine pyodermas. *Amer. J. vet. Res.* 1986, *47*, 229-231.
- Muller GH, Kirk RW, Scott DW*: Small animal dermatology. 4th ed. WB Saunders Company, Philadelphia, 1989.
- Noble WC, Kent LF*: Antibiotic resistance in *Staphylococcus intermedius* isolated from cases of pyoderma in the dog. *Vet. Dermatol.* 1992, *3*, 71-74.
- Phillips WE Jr, Kloos WE*: Identification of coagulase-positive *Staphylococcus intermedius* and *Staphylococcus hyicus* subsp. *hyicus* isolates from veterinary clinical specimens. *J. clin. Microbiol.* 1981, *14*, 671-673.
- Quadros E*: Furunculosis in dogs. Aetiology, pathogenesis and treatment. A clinical study. *Acta vet. Scand.* 1974, suppl. 52.
- Raus J, Love DN*: Characterization of coagulase-positive *Staphylococcus intermedius* and *Staphylococcus aureus* isolated from veterinary clinical specimens. *J. clin. Microbiol.* 1983, *18*, 789-792.
- Schleifer KH*: Gram-positive cocci. In: *Sneath PHA, Mair NS, Sharpe ME* (ed.): Bergey's manual of systematic bacteriology, vol. 2, pp. 999-1103. Williams & Wilkins, Baltimore, 1986.
- Southern EM*: Detection of specific sequences among DNA fragments separated by gel electrophoresis. *J. mol. Biol.* 1975, *98*, 503-517.
- Talan DA, Staatz D, Staatz A, Goldstein EJ, Singer*

K, Overturf GD: *Staphylococcus intermedius* in canine gingiva and canine-inflicted human wound infections: laboratory characterization of a newly recognized zoonotic pathogen. J. clin. Microbiol. 1989a, 27, 78-81.

Talan DA, Staats D, Staats A, Overturf GD: Frequency of *Staphylococcus intermedius* as human nasopharyngeal flora. J. clin. Microbiol. 1989b, 27, 2393.

Wegener HC, Pedersen K: Variations in antibiograms and plasmid profiles among multiple isolates of *Staphylococcus intermedius* from pyoderma in dogs. Acta vet. Scand. 1992, 33, 391-394.

Woldehiwet Z, Jones JJ: Species distribution of coagulase positive staphylococci isolated from dogs. Vet. Rec. 1990, 126, 485.

### Sammendrag

*Antibiogrammer og ribotyper blandt Staphylococcus intermedius isoleret fra raske hunde sammenlignet med isolater fra hunde med otitis externa eller furunkulose.*

I alt 60 *Staphylococcus intermedius* stammer, 15 fra raske hunde, 9 med otitis externa, og 36 med furun-

kulose blev ribotypet (60 stammer) og undersøgt for følsomhed overfor forskellige antibiotika (50 stammer). Tres procent af de 50 resistensundersøgte isolater udviste resistens overfor penicillin, 24% overfor spiramycin, 20% overfor tetracyclin, 16% overfor chloramphenicol og 2% overfor fucidin. Alle isolater var følsomme overfor amoxycillin med clavulansyre, enrofloxacin og sulfonamider med trimethoprim. Der var ingen signifikant forskel på resistensmønstrene mellem isolater fra de 3 grupper hunde. Hos de 60 ribotypede stammer blev 10 forskellige ribotyper påvist: 6 blandt isolater fra otitis, 6 fra furunkulose og 5 fra raske hunde. En af ribotyperne (profil C) dominerede blandt isolater fra raske hunde, mens en anden ribotype (profil A) dominerede blandt isolater fra hunde med pyodermi. Profil A blev ikke påvist hos nogen af stammerne fra de raske hunde. Hos 5 forskellige hunde med pyodermi var i en tidligere undersøgelse påvist to forskellige kloner baseret på plasmidprofiler og antibiogrammer. Heraf tilhørte den ene klon ifølge denne undersøgelse altid ribotype A. Dette antyder, at der i relation til furunkulose findes distinkte subpopulationer af *S. intermedius*, som udviser forskellig patogenitet over for hunde.

(Received December 24, 1994; accepted May 24, 1995).

Reprints may be obtained from: K. Pedersen, Department of Veterinary Microbiology, The Royal Veterinary and Agricultural University, Bülowsvej 13, DK-1870 Frederiksberg C, Denmark.