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A literature study on Methicillin-resistant *Staphylococcus pseudintermedius* (MRSP): classification, resistance mechanisms, risk factors and clonal distribution.

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En litteraturstudie om methicillinresistent *Staphylococcus pseudintermedius* (MRSP): klassificering, resistensmekanismer, riskfaktorer och klonal distribution

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ABSTRACT

The increased frequency of reported Methicillin-resistant *Staphylococcus pseudintermedius* (MRSP) is of great concern to small animal healthcare due to limited options of antimicrobial therapy. The aim of this literature study is to investigate 1) characteristics and classification of *S. pseudintermedius,* 2) resistance mechanisms and emergence of MRSP, 3) risk factors for selection of MRSP, and 4) clonal distribution around the world. *S. pseudintermedius* is a skin and mucosal commensal in healthy dogs and cats, but can cause infections, including pyoderma.

Beta-lactam antibiotics inhibit bacterial growth by binding to the penicillin binding protein (PBP), a vital enzyme in the synthesis of the bacterial cell wall. Methicillin-resistance in *S. pseudintemedius* is encoded by the *mecA* gene, that expresses modified PBP, named PBP2a or PBP2', with a lower affinity for beta-lactams. The *mecA* gene is located on the staphylococcal chromosomal cassette, a mobile genetic element. In addition to beta-lactams, MRSP can be resistant to other classes of antibiotics drugs.

The main risk factors for selection of MRSP are hospitalization, veterinary visits and former treatment with antibiotics. MRSP infection is therefore considered to be a nosocomial infection. The major clonal lineage predominant in Europe is ST71-J-t02-II–III; whilst in North America it is ST68-C-t06-V. Colonization of *S. pseudintermedius* in human is rare, and only one case of MRSP has been found up to date. Resistance genes are believed to be shared between species of staphylococci by horizontal gene transfer. However, more studies are needed to fully understand the mechanisms of horizontal gene transfer of the *mecA* gene.

SAMMANFATTNING

Ett växande problem inom smådjursveterinärmedicinen är methicillinresistent *Staphylococcus pseudintermedius* (MRSP) som begränsar valet av antibiotikum vid behandling. Syftet med denna litteraturstudie är att kartlägga 1) egenskaper och klassificering av *S. pseudintermedius,* 2) resistensmekanismer och uppkomst av MRSP, 3) riskfaktorer som selekterar för MRSP och 4) den klonala distributionen i världen. *S. pseudintermedius* är normalflora på hud och slemhinnor hos friska hundar och katter, men kan även orsaka opportunistiska infektioner, bl.a. pyodermi.

Antibiotikaklassen beta-laktamer utövar sin antibakteriella effekt genom att inhibera penicillinbindande protein (PBP), ett enzym involverad i cellväggssynten. Methicillinresistensen hos MRSP kodas av m*ecA*-genen, som ger upphov till modifierat PBP, kallat PBP2a eller PBP2', med en lägre affinitet för beta-laktamer. *MecA* genen är belägen på stafylokockens kromosomkassett, ett mobilt genelement. MRSP kan vara resistenta mot andra antibiotikaklasser utöver beta-laktamer.

De riskfaktorer som selekterar för MRSP är sjukhusvistelse, veterinärbesök och tidigare antibiotikabehandling. Därför anses MRSP-infektioner vara nosokomiala smittor. Den klon som dominerar i Europa är ST71-J-t02-II–III, och ST68-C-t06-V dominerar i Nordamerika. *S. pseudintermedius* koloniserar sällan människor, och endast ett fall av MRSP hos människa har rapporterats. Olika arter av stafylokocker tros dela resistensgener genom horisontell genöverföring. Dock behövs fler studier för att klarlägga mekanismer för horisontell genöverföring av *mecA-genen* mellan stafylokocker.

INTRODUCTION

The rate and extent of antimicrobial resistance in bacteria has increased in recent years. The World Health Organization (WHO) defines antimicrobial resistance as the "…resistance of a microorganism to an antimicrobial drug that was originally effective for treatment of infections caused by it" (WHO, 2014). The use of antimicrobial drugs selects for resistant strains of bacteria already present in the population, rather than actually inducing resistance (Hirsh *et al*., 2004; Quinn *et al*., 2011).

Since the first description of *Staphylococcus pseudintermedius* as a novel species in 2005 (Devriese *et al*., 2005), methicillin-resistant *Staphylococcus pseudintermedius* (MRSP) was reported with increased frequency in the following year (van Duijkeren *et al*., 2011). The bacterium is both a commensal on dogs, and a pathogen causing canine pyoderma and wound infections (van Duijkeren *et al*., 2011). Moreover, MRSP are often resistant to several different classes of antibiotics (Perreten *et al*., 2010). This limits the treatment options and is considered a threat to small animal therapy in veterinary medicine (van Duijkeren *et al*., 2011).

This literature study aims to investigate the following:

1) Characteristics and classification of *S. pseudintermedius*
2) Emergence of MRSP and resistance mechanisms
3) Risk factors for selection of MRSP
4) Clonal distribution around the world

MATERIALS AND METHODS

Databases used were PubMed, Web of Science and Primo.

The main search terms were (MRSP OR "Methicillin-resistant *staphylococcus pseudintermedius*"), (antibio* OR antimicr*), (dog OR Canine), geneti*, prevalence, SCC, *mecA* and horizontal gene transfer.

LITERATURE STUDY

What is antimicrobial resistance?

Resistance to antimicrobial drugs can occur due to acquired resistance, either by mutation in the chromosomal DNA, or by acquiring genetic material. Chromosomal mutation often leads to a change in the bacterial structure, e.g. altered antimicrobial target proteins. These rare, spontaneous changes in the bacterial DNA are due to mistakes during DNA replication, and are not caused by the presence of antibiotics. The mutations can be either beneficial, in this case by causing antimicrobial resistance, or disadvantageous to the survival of the bacteria, even lethal. Mutations in genes leading to expression of efflux proteins can induce multiple antibiotic resistances. (Hirsh *et al*., 2004; Quinn *et al.*, 2011)

The mechanism of more importance to the development of antibiotic resistance is the exchange of transferable DNA structures, usually plasmids. These mobile gene elements carrying resistance genes can be spread to other bacteria by (see Figure 1):

- Transduction – DNA incorporated in bacterial phages (virus that infects bacteria) and transferred between bacteria by phages.
- Conjugation – The transfer of plasmid or chromosomal DNA from donor to recipient through a sex pilus.
- Transformation – The ability of certain bacteria genera to pick up naked DNA from the environment.
- Transposition – Gene segments can change position within the bacterial genom.
- Integrons – Consists of an intergrase gene that can capture gene cassettes coding for antibiotic resistance (Hirsh *et al*., 2004; Quinn *et al.*, 2011).

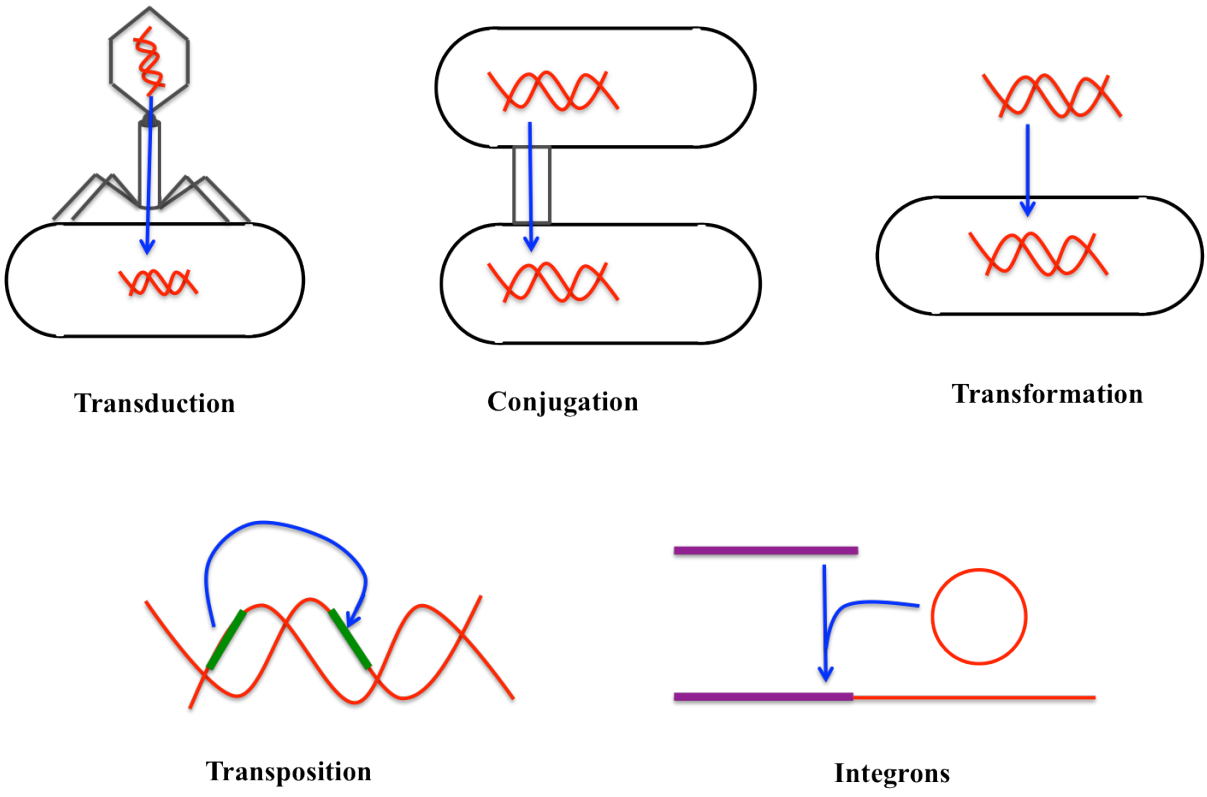

Figure 1. Mobile gene elements. By Yusi Fang modified from (2013MMG320B, 2013).

Staphylococcus pseudintermedius – Characteristics and classification

While *S. aureus* is the predominant *Staphylococcus* species colonizing and infecting humans (Sung *et al*., 2008), *S. pseudintermedius* is the more important pathogen in dogs (van Duijkeren *et al.*, 2011).

S. pseudintermedius is a coagulase-positive facultative anaerobe gram-positive coccus. It appears as small, round, light grey, opaque colonies with a diameter of 1–2 mm when cultured. The bacteria show a double zone of haemolysis on bovine blood agar: an inner zone of complete haemolysis caused by α-haemolysin, and an outer zone of partial haemolysis caused by β-haemolysin. The bacteria are catalase-positive and oxidase-negative. Their micromorphology is cocci arranged in groups (VetBact, 2014).

S. pseudintermedius is a commensal on skin and mucosa of healthy dogs and cats, and can be isolated from the nares, mouth, anus, groin and forehead (Kadlec & Schwarz, 2012). In rare cases, the bacteria have been found in humans and horses (Kadlec & Schwarz, 2012). As a pathogen, it can cause opportunistic infections on the skin, including pyoderma, otitis externa, wound infections, abscesses, infections on other body tissues and cavities, and post-operative wound infections (Weese & van Duijkeren, 2010; van Duijkeren *et al*., 2011). Kawakami *et al*. (2010) state that *S. pseudintermedius* is considered to be the most important pathogen causing canine pyoderma.

S. intermedius was first described in the 1970s based on phenotypical characteristics. In 2005, *S. pseudintermedius* was discovered as a novel species using the technique of 16S rRNA gene sequence analysis. *S. intermedius* was reclassified into three closely related clusters: *S. intermedius, S. pseudintermedius* and *S. delphini,* which together make up the *Staphylococcus intermedius* group (SIG) (Sasaki *et al*., 2007). The different species within the SIG group are difficult to distinguish by using standardized routine diagnostic methods only. Since *S. pseudintermedius* is the cluster that colonizes and infects dogs, it is recommended to classify all strains of the SIG group found on dogs as *S. pseudintermedius,* if molecular analysis has not been carried out. (Weese & van Duijkeren, 2010; van Duijkeren *et al.,* 2011)

Resistance to beta-lactams - mechanisms

Beta-lactam antibiotics inhibit bacterial growth by binding to the penicillin binding protein (PBP), a vital enzyme in the synthesis of the bacterial cell wall (Weese & van Duijkeren, 2010). In the 1980s, McDougal and Thornsberry (1986) found that staphylococci started to show resistance against penicillin and cephalosporin. The mechanism behind the resistance was that the enzyme beta-lactamase produced by some strains of staphylococci were able to hydrolyse and thereby inactivate beta-lactam antibiotics. Beta-lactamase inhibitors, e.g. clavulanic acid in combination with beta-lactam antibiotics, have shown to have some effect on beta-lactamase producing strains (McDougal & Thornsberry, 1986).

Methicillin-resistant *Staphylococcus aureus* (MRSA) and MRSP show resistance via a different mechanism in addition to beta-lactamase production, by expressing a modified PBP, known as PBP2a or PBP2', with a lower affinity for beta-lactams. The antibiotics cannot inhibit the enzyme and fail to exercise its inhibitory effect on bacterial growth (Hanssen &

Sollid, 2006; Weese & van Duijkeren, 2010). MRSP has become a significant problem worldwide due to the limited amount of antibiotic for the treatment of *S. pseudintermedius* infections (Solyman *et al.,* 2013).

Methicillin-resistance in *S. pseudintemedius* is encoded by the *mecA* gene. It is located on the staphylococcal chromosomal cassette (SCC). The different types of SCCmec elements found in MRSP are SCCmec II-III, SCCmec III, SCCmec IV, SCCmec V, SCCmec VII and non-typeable cassettes (van Duijkeren *et al.,* 2011).

Subtyping

Other methods in addition to SCCmec typing are used for subtyping, identification and classification of MRSP. Multilocus sequence typing (MLST) is a method used to identify sequence types (designated ST) and hence the genetic structure, as well as tracking the clonal distribution of MRSP (Solyman *et al.,* 2013). Typing scheme for staphylococcal protein A (*spa*) gene (designated t) and pulse-field gel electrophoresis (PFGE) are used to compare isolates (Ruscher *et al*., 2010; van Duijkeren *et al*., 2011).

Resistance to non-betalactams

MRSP are considered to be resistant to all beta-lactam antibiotics. Many isolates also show resistance against other classes of antibiotics, e.g. tetracyclines, macrolides, lincosamides, aminoglycosides, trimethoprim, chloramphenicol and fluoroquinolones (Perreten *et al.,* 2010; Weese & van Duijkeren, 2010; van Duijkeren *et al.*, 2011; Cain, 2013). Different resistance genes mediate different resistance mechanisms, including membrane-associated efflux, altered target molecules, enzyme inactivation, protection proteins and altered membrane channel permeability (Perreten *et al*., 2010; Weese & van Duijkeren, 2010; van Duijkeren *et al.*, 2011; Cain, 2013).

MIC and antibacterial susceptibility test

Minimal inhibitory concentration (MIC) is the lowest concentration of an antibacterial agent that inhibits growth of a bacterial isolate (Quinn *et al.,* 2011).

Broth dilution, disc diffusion and agar gradient are different tests to determine susceptibility of a bacterial isolate to different antibiotics in vitro. In broth dilution, a standard amount of bacteria is added to broth with varying or two-fold dilution of antibiotic concentrations. Broth dilution is considered to be an accurate method in finding the lowest concentration of an antibiotic that inhibits the growth of the tested bacteria. (Engelkirk & Duben-Engelkirk, 2008; Quinn *et al*., 2011)

The commonly used Kirby-Bauer disc diffusion method is a relatively simple and economical method, most suitable for testing rapidly aerobic bacteria. The bacterial sample is spread on an agar plate and antimicrobial discs with specific amounts of antibiotics are placed on the inoculated surface. After incubation, the diameter of the inhibition zone of each antibiotic is measured in millimetres and compared to international standards. (Quinn *et al.,* 2011)

The Clinical and Laboratory Standards Institute (CLSI) recommends using a breakpoint for oxacillin (a beta-lactam antibiotic) susceptibility of *S. pseudintermedius* equivalent to the breakpoint of *S. aureus*, a value >2 mg/l (Bemis *et al.,* 2009; A. Bergström *et al.,* 2012). However, a study by Bemis *et al*. (2009) concluded that the criteria stated by CLSI were not sufficient to detect all *mecA*-associated resistance in *S. pseudintermedius*. Instead, oxacillin disk diffusion zone diameters of ≤17 mm and a oxacillin MIC of ≥ 0.5 μg/ml is found to accurately determine oxacillin resistance in *S. pseudintermedius* mediated by the *mecA* gene.

The emergence of MRSP and horizontal gene transfer (HGT)

Methicillin-resistant canine staphylococci were first isolated in the 1980s by Pellerin *et al*., (1998) in France. The bacteria were reported to be resistant to penicillin, oxacillin and cloxacillin. In 1999, Gortel *et al.* (1999) suggested that this resistance was mediated by the *mecA* gene, since isolates possessing the *mecA* gene were more likely to be resistant to several kinds of beta-lactam antibiotics than isolates lacking the *mecA* gene. The *mecA* gene is located on the mobile genetic element SCCmec. Since the first reports, MRSP has been reported with increased frequency (Weese & van Duijkeren, 2010).

Bannoehr *et al.* (2007) studied 105 isolates of the SIG group, obtained from both healthy and diseased animals from several different countries around the world. By sequencing the *mecA* gene, they found in their study that the *mecA* genes of *S. pseudintermedius* were closely matched with *mecA* genes in *S. aureus*. A recent study by McCarthy *et al.* (2014) investigated the genomic background of 12 *S. pseudintermedius* isolates with varying resistance degree from several different countries, including the UK and Germany. The authors suggested that resistance was spread amongst *S. pseudintermedius* through horizontal gene transfer (HGT) with mobile genetic elements. Resistance to beta-lactams was not likely to be mediated by plasmid conjugation since the plasmids in the investigated strains did not carry any resistance genes. Instead, the authors concluded that HGT is most likely to be mediated by transposons and bacteriophage transduction. The SCCmec element of *S. pseudintermedius* was highly homologous with the ones of *S. aureus* and coagulase negative staphylococcus (CoNS), such as *S. epidermidis* and *S. haeomlyticus* (McCarthy *et al.*, 2014). Together with the study by Bannoehr *et al.* (2007), this supports the hypothesis of HGT between species of *staphylococci*.

However, Wang *et al.* (2012) do not believe SCCmec of *S. pseudintermedius* are acquired from *S. aureus*, since the SCCmec type in their study differed from the SCCmec types isolated from Methicillin resistant *Staphylococcus aureus* in their previous study (Zhang *et al.,* 2011).

Major risk factors affecting selection of MRSP

A recent case-control study by Lehner *et al*. (2014) examined 20 different variables of risk factors for MRSP in dogs and cats in Germany. Samples were taken from a university teaching hospital and non-university teaching clinics. The disc diffusion method was used to determine antimicrobial resistance. In the univariable regression analysis, 11 variables were found to significantly increase the odds ratio of MRSP. However in the multivariable logistic regression analysis, the findings of interest were that animals that were hospitalized or

made frequent visits to a veterinarian had a higher risk of carrying MRSP. Also, the prevalence of MRSP was higher in the university teaching hospital (87/360, 24.2 %) than the non-university veterinary clinics (52/680, 7.7 %). The authors concluded that MRSP is to be considered a nosocomial (healthcare-associated) infection, where hospitalization and frequent veterinary visit are the major risk factor for MRSP infection. A study by Nienhoff *et al.* (2011) including 814 dogs found treatment with antibiotics as a main risk factor, in addition to hospitalization and recent veterinary contact.

Loeffler *et al.* (2007) studied 12 patients with recurrent pyoderma or otitis that had been treated with systemic beta-lactams or fluoroquinolones for bacterial infection. *S. intermedius* resistant to at least 5 antimicrobial classes was found, 11 isolates were resistant to antibiotics licensed for systemic use in most European countries. This study also suggests that the use of antibiotics during a long period of time select for multiresistant *S. intermedius*.

Clonal distribution of MRSP around the world

An international multicentre study by Perreten *et al.* (2010), aimed to map the phenotypic and genotypic resistance profiles of MRSP colonizing dogs in North America and several different European countries. The study included 103 isolates of MRSP from healthy dogs and dogs with clinical symptoms. The authors found that ST71-J-t02-II–III was the clonal MRSP lineage that was predominant in Europe, whilst in North America it was the ST68-C-t06-V clonal lineage. All isolates were found to be carrying the *mecA* gene and were resistant to oxacillin. Both clones were also resistant to several other antibiotic classes, by different resistance genes. Some MRSP isolates belonged to unique sequence types (ST69, ST113, ST114 and ST116). Haenni *et al*. (2014) found the ST71–t02–II-III clone in France, the same European clone described by Perreten *et al.* (2010). Another study, in Germany and several other European countries, found MRSP isolates belonging to the sequence type ST71 and the majority harboured spa type t02 (Ruscher *et al*., 2010). However, in this study SCCmec III was found to be the most prevalent SCCmec type, differing from the findings of Haenni *et al.* (2014) and Perreten *et al.* (2010).

Wang *et al*. (2012) were the first to investigate MRSP from canine pyoderma in China and identified ST71-t06-II-III as the most prevalent genotype. A total of 260 pyoderma samples were taken from dogs in northern China. Broth dilution was carried out to determine antibiotic susceptibility. Thirty-three isolates were identified as MRSP, all harbouring the *mecA* gene encoding resistance against oxacillin. In addition, all isolates were resistant to tetracycline, erythromycin (macrolide) and clindamycin (lincosamide). Wang *et al.* found the same ST type (ST71) as the European ST71-J-t02-II–III clone found by Perreten *et al. (*2010), but with a different *spa* type (t06 in China and t02 in Europe).

Zoonotic aspects of MRSP

Colonization of *S. pseudintermedius* is very rare in humans unlike *S. aureus*. Therefore, the importance of MRSP as a zoonotic agent is not as great as that of MRSA. However, *S. pseudintermedius* is found more often in owners of dogs with deep pyoderma. The colonization is usually transient, since humans are not natural hosts of the bacteria. (van Duijkeren *et al*., 2011)

Stegmann *et al*. (2010) were the first to report a case of human infection with MRSP. The isolate was identified as ST71-t02-J with SCCmec II–III, the sequence type circulating among dogs in Europe. The patient was the owner of a dog, which needed home care and underwent several antibiotic treatments. The dog was euthanized and samples could not be obtained, and thus it was not possible to determine the source of infection. However, transmission of MRSP from the dog to its owner is a possibility. Stegmann *et al.* (2010) regard the zoonotic aspect of MRSP as an increasing problem, and think that "last resort antibiotics" as treatment for methicillin-resistant staphylococci should be saved for human use, rather than treatment of animals.

S. pseudintermedius has some importance in dog-bite wounds were it can be an opportunistic pathogen. *S. pseudintermedius* can be misidentified as *S. aureus* in dog-bite wounds on humans and the prevalence may therefore be underreported. (Weese & van Duijkeren, 2010)

DISCUSSION

As MRSA is considered to be a major nosocomial pathogen amongst humans worldwide (Graffunder & Venezia, 2002), MRSP could be the corresponding threat amongst dogs in veterinary medicine. Lehner *et al.* (2014) and Nienhoff *et al*. (2011) found significant correlation between MRSP infection and hospitalization and veterinary visits. One reason could be the direct transmission between dogs in the veterinary clinics, since dogs are found to carry MRSP for at least six months after acquisition (A. Bergström *et al.,* 2012). Another reason is the possible risk of indirect transmission between dogs via environmental surfaces in hospitals. Singh *et al.* (2013) aimed to determine personnel clothing as a risk factor for the spread of methicillin-resistant staphylococci. Correlation was found for MRSA but not MRSP. However, the study included a very small sample size (n=4 in the group with MRSA) and it is therefore hard to draw any definite conclusion.

In an outbreak of MRSA infection among horses in a Swedish veterinary hospital, 11% of the environmental samples, from walls, floor, treatment areas, doorknobs etc. were positive for MRSA, and the hospital environment can therefore be considered a major risk factor for indirect transmission of MRSA (K. Bergström *et al*., 2012). The fact that a similar correlation has not yet found between MRSP and environmental contamination should not exclude the possibility of this being a risk factor, since the two species of staphylococci share many characteristics. Anyhow, more emphasis should be put on the importance of isolation of infected individuals and hygiene demands of staff, equipment and environment.

Antibiotic use should also be considered as a risk factor, since it selects for resistant bacteria. Both Nienhoff *et al*. (2011) and Loeffler *et al.* (2007) found significant correlations between

antibiotics treatment and the carriage of MRSP. In Sweden, it is recommended to avoid antibiotic treatment of patients with MRSP to reduce the risk of further development of resistance (SVA, 2014). However, there are difficulties in standardizing the studies for comparison between different countries, due to many factors involved, e.g. antibiotics available in the country, local legislation, personal experience and preferences affect the use and choice of antibiotics.

Solyman *et al.* (2013) believe that the understanding of clonal distribution of MRSP around the world provides great possibilities for control programs to reduce the spread. The information is also useful for the development of new treatment methods, e.g. vaccines or phage therapy that can target the specific clone present.

Perreten *et al*. (2010) found the European ST71-t06-II-III clone to be multi-resistant to several classes of antibiotics, and to have harboured more resistance genes than other minor sequence types. The results of Perreten *et al.,* (2010) and Wang *et al*. (2012) show ST71 to be a successful MRSP type spread around large parts of the world. Since Lehner *et al*. (2014), Nienhoff *et al.* (2011) and Loeffler *et al*. (2007) found hospitalization, veterinary visits and antibiotic treatment to be the major risk factors for infections with MRSP, it is likely that selection for resistance takes place in the hospital environment. In the presence of these risk factors and a selection pressure, the MRSP are more likely to acquire and harbour other resistance genes. Although carrying excess genes is costly for the bacterium, it is advantageous in such environments. Therefore, the predominant clones become more successful and harder to eradicate.

There are several studies examining the clonal distribution of MRSP around the world, but studies on the mechanism of spread have not been found. The *spa* types and SCCmec types differ in location, but the same ST71 lineage is found in both Europe and East Asia, suggesting that ST71 is the most successful ST type in dogs. Since dogs do not travel to the same extent as humans, the *mecA* gene could possibly be carried to new geographic regions by human *S. aureus*. McCarthy *et al.* (2014) and Bannoehr *et al*. (2007) found the SCCmec element of S. *pseudintemedius* to highly match the SCCmec elements of *S. aureus* and other species of staphylococci, and thus believe in HGT between staphylococcal species, supporting the hypothesis of human carriage. However, both studies included samples from various different regions around the world, making it hard to draw certain conclusions.

Wang *et al.* (2012) on the other hand present a view that differ from the ones presented in other studies (Bannoehr *et al*., 2007; McCarthy *et al.*, 2014). Wang et al. compared the SCCmec type of MRSP found in canine patients with their previous study carried out in the same regions by Zhang *et al*. (2011) on SCCmec in *S. aureus.* They identified differences in SCCmec and concluded that SCCmec has not been acquired from *S. aureus.* However, the sample size was rather small (n=33, Wang *et al*., n=18, Zhang *et al.*). One can therefore question whether such conclusions can be drawn from the results. Also, the study population was different where Zhang *et al*. took samples from dogs, cats and veterinary staff (not specified whether healthy or diseased individuals), making the two studies incomparable. Moreover, the aim of the study by Zhang *et* al. was to investigate the occurrence of MRSA

in pets and veterinary staff, and not comparisons in SCCmec. Therefore, the different study designs of the two studies also make them less comparable. One should also bear in mind that the fact that HGT has not yet been found does not necessarily imply it not occurring.

Loeffler *et al.* (2007) claim to have studied *S. intermedius*. However, only phenotypic speciation and not molecular analysis was carried out to identify the staphylococcal isolates as *S. intermedius*. Besides, at the beginning of the study 18 months earlier, *S. pseudintermedius* was just described as a novel species belonging to the SIG group together with *S. intermedius* and *S. delphini*. Since *S. pseudintermedius* is the most frequent species of the SIG group to colonize dogs, old reports on *S. intermedius* are considered to belong to the species *S. pseudintermedius* (Weese & van Duijkeren, 2010).

That MRSP is becoming an increasing problem in veterinary medicine is evident, but its zoonotic role is not yet fully understood. The zoonotic aspect of MRSP is certainly of less importance than that of MRSA, since even colonization of *S. pseudintermedius* in humans is rare. One human case of MRSP has been reported, but it was not possible to say whether the infection came from an animal or not. However, if resistance genes can be acquired by human commensal *S. aureus* from canine MRSP, the zoonotic problem will become a serious threat to human. On the other hand, zoonoses can go both ways where humans can act as a source of infection for dogs. The selection pressure in human medicine is even greater due to the more extensive use of antibiotics. In some countries, certain antibiotics are restricted to human medicine (meaning that they are not available for the treatment of sick animals) and hence the problem in veterinary medicine may become more urgent. In any case, further studies are needed to investigate and fully understand the horizontal gene transfer of *S. pseudintermedius* SCCmec, e.g. by starting to study the mechanisms *in vitro*.

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