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Methicillin-resistant *Staphylococcus aureus* in people living and working in pig farms

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SUMMARY

We compared the prevalence of human and animal methicillin-resistant *Staphylococcus aureus* (MRSA) at pig farms in The Netherlands, and related this to individual and farm-level characteristics. More than half of the farms investigated (28/50) had MRSA in pigs or stable dust and about one third (15/50) of person(s) were identified as MRSA carriers. Human carriage was found only on farms with MRSA-positive pigs or dust. MRSA strains in human samples were the same *spa*-type as found in pigs and all were not typable by pulsed-field gel electrophoresis (NT-MRSA). Multivariate analyses showed that risk factors for human MRSA carriage were: working in pig stables (OR 40, 95% CI 8–209) and the presence of sows and finishing pigs (OR 9, 95% CI 3–30). Veterinary sample collectors sampling the pigs showed transient MRSA carriage only during the day of the farm visit. Working in pig stables with MRSA-positive pigs poses a high risk for acquiring MRSA, increasingly so when contact with live pigs is more intensive or long lasting.

Key words: Domestic pigs, MRSA, prevalence, risk factors, *Staphylococcus aureus*, zoonoses.

INTRODUCTION

The antimicrobial agent methicillin was introduced into clinical practice in 1960 and methicillin-resistant *Staphylococcus aureus* (MRSA) was described 1 year later [1]. Worldwide, MRSA is now responsible for considerable mortality, morbidity and health-care expenditure both in hospital and community settings [2, 3]. In The Netherlands MRSA has been controlled effectively in hospitals by a search and destroy strategy [4]. In 2006, 2% of the *S. aureus* invasive isolates

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in The Netherlands were resistant to methicillin, compared to 20–40% in surrounding countries [5].

The Netherlands was the first country to report patients with a specific MRSA variant associated with pigs [6]. This strain was not typable by pulsed-field gel electrophoresis (PFGE) and was therefore referred to as NT-MRSA [7]. Since then an increasing number of Dutch studies have reported NT-MRSA in people in contact with pigs, i.e. pig farmers and family, co-workers and veterinarians [8–11]. In a pilot survey, Voss *et al*. [6] found a 23% prevalence of NT-MRSA in 26 pig farmers. A study of pigs in slaughterhouses revealed that more than one third of them carried NT-MRSA [12]. Cattle (veal) farming was also identified as a risk factor for NT-MRSA carriage in humans [13, 14].

Since mid-2006, pig farmers and their family members are screened for MRSA at ambulatory care or before hospitalization. As a result thereof, the proportion of NT-MRSA in the national surveillance increased by 20% in the second half of 2006 and by 30% in 2007 [13] (unpublished data, RIVM) and even a threefold increase was reported from a hospital in the south of the country [15]. Up to now, NT-MRSA has been found in France, Germany, Austria, Denmark, Belgium and Canada [16–21].

Despite published reports, no representative estimate of the prevalence of NT-MRSA on Dutch farms is available. Our objective was to obtain further insight into the prevalence and determinants of human carriage of NT-MRSA in relation to the presence of NT-MRSA in pigs on the farm.

MATERIALS AND METHODS

Selection of farms

A cross-sectional prevalence survey was conducted during the period January–October 2007. Farms were randomly selected from a complete list of pig farms in The Netherlands. On 50 farms we expected to include 140 persons, i.e. 50 farmers and their family (average Dutch family size 2·8), sufficient to estimate the MRSA point-prevalence, assuming 23% positivity [6], accepting a 5% risk of type I error with a precision of 10% and a design effect of 2 [anticipating clustering of cases; calculation with Epi-Info 6.04 (CDC, Atlanta, GA, USA)]. The study protocol was approved by the medical ethical committee of the Therapeutic Drug Evaluation Foundation (STEG, Almere, The Netherlands).

Sampling and questionnaires at pig farms

Procedures for human subjects

Written informed consent to participate in the research was obtained; for children aged <18 years parental consent was requested. A short questionnaire and nasal swabs (from both anterior nares) were taken by the research assistant visiting the farm. The questionnaire addressed individual factors such as age, sex, intensity of contact with pigs or other animals, potential other risk factors for MRSA carriage and self-reported medical history related to MRSA infection or (skin) problems.

Research assistants took their own nasal swabs just before they visited the farm, immediately after and the morning following the visit.

Procedures for animal data

Data on farm-related factors included size and type of farm, age groups of pigs present (sows, suckling piglets, weaned piglets, gilts and finishing pigs), measures of hygiene implemented, feeding method and housing characteristics. Pigs were sampled by nasal swabs and 60 samples were collected per farm, representative for the age group(s) of pigs present. These swabs were pooled into 10 pools of six swabs. Additionally, at each farm five environmental samples of dust were collected by wiping the top of the pen separations in different compartments of the pig-houses. Visits were separated by at least 1 day and limited to two farms per week.

Laboratory analysis

Samples were cultured at the hospital laboratory in Breda (human samples) and the RIVM laboratory (pig and dust samples). Samples were first enriched in Mueller–Hinton broth with 6·5% NaCl, followed by selective enrichment in Phenol Red mannitol broth with 75 mg/l aztreonam and 4 mg/l oxacillin or ceftizoxime. After culturing on sheep blood agar and MRSA screen agar (Oxoid, Basingstoke, Hants, UK), suspected colonies were confirmed by polymerase chain reaction (PCR) for the *S. aureus*-specific DNA fragment [22], the *mecA* gene [23] and the Panton Valentine leukocidin toxin genes [24].

For the human isolates, methicillin resistance was screened by a disk diffusion test using a cefoxitin disk and confirmed by the presence of the *mecA* gene by PCR. All MRSA strains were typed by *spa* typing [25] and strains in human samples not yet identified as

NT-MRSA were checked for typability by PFGE [26]. Furthermore, in human samples, susceptibility was determined for 21 antimicrobial agents with the VITEK system (bioMérieux SA, Craponne, France) according to the manufacturer's instructions.

Data analysis

Data were entered into Access, double-checked and verified with questionnaires and analysed with SPSS version 15.0 (SPSS Inc., Chicago, IL, USA) and SAS version 9.1 (SAS Institute, Cary, NC, USA). A dataset was created with individual human results and aggregated animal and farm data. Summary variables were created, such as a score for 'personal hygiene in the stable' based on the following hygiene measures: separate entrance and exit, changing room, showers, water, soap, boots, overalls and disinfection footbath.

The relationship between MRSA presence in human and animal and/or dust samples was investigated by χ^2 test. Risk factors for MRSA carriage were first identified by univariate logistic regression analysis. For further analyses we considered exposure to MRSA-positive pigs or dust a prerequisite or 'necessary cause' for MRSA positivity in humans and therefore further analyses were limited to data from farms where pigs or dust tested positive. Multivariate regression analysis by stepwise, forward entry included factors influencing MRSA positivity in humans ($P<0{\cdot}2$ in univariate analysis) and logical interaction factors thereof. A random cluster effect was included in the model to adjust for the fact that observations of humans on the same farm might not be independent.

Spa-types of human and animal MRSA samples were compared for each farm, including the results of the samples taken from the sample collectors pre- and post-farm visit. Antibiotic resistance patterns of human MRSA strains were compared for each *spa*-type by χ^2 analysis.

RESULTS

Altogether 106 farms were contacted until 50 farmers (47%) agreed to participate in the research. The response rate was similar in different regions and types of pig farms. The most common reasons for non-participation were no interest or time, or retirement from farming. A total of 232 people were sampled on 50 farms: 50 farmers, 171 family members and 11 co-workers. The individual participation on the selected farms was high: we collected swabs and questionnaires from 221/231 reported household members (96%).

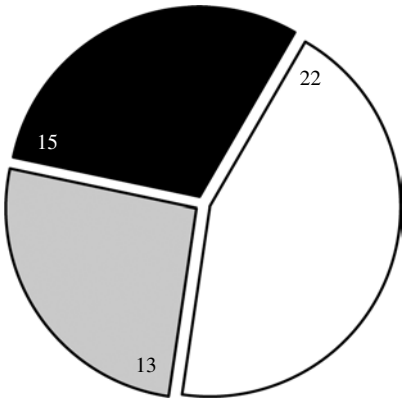

Fig. 1. Prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) per farm. Number of farms (total 50) indicated in segments. Farms with one or more MRSA-positive persons and MRSA in pigs or dust (black area); farms with MRSA in pigs or dust only (grey); farms completely MRSA negative (white).

MRSA status per farm

On 28/50 (56%) of the farms pig or dust samples tested positive (hereafter referred to as 'MRSA-positive farms') and on 15/50 (30%) of the farms one or more MRSA-positive persons were found. MRSA in humans was only found on MRSA-positive farms (Fig. 1). On the 15 farms with MRSA-positive people, pigs were also positive with the exception of one farm; on this farm, however, dust samples were positive. The prevalence of MRSA in farm residents was the highest on farms with both sows and finisher pigs. Finishing farms were more often MRSA positive than farms with sows only, but only part of the positive finishing farms (38%) housed MRSA-positive people while on all MRSA-positive farms where sows were present, MRSA was also found in farm residents (see Fig. 2).

MRSA status per person – univariate analysis

MRSA was identified in 33/232 people (14%). Table 1 shows the relationship between MRSA positivity and the main potential risk factors (univariate analysis). At the individual level, living on a MRSA-positive farm was the most important risk factor. The higher the proportion of positive samples from pigs or dust per farm, the higher the positivity rate in people. The

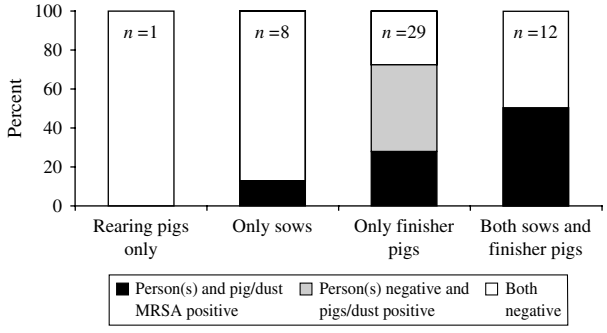

Fig. 2. Proportion of farms with methicillin-resistant *Staphylococcus aureus* (MRSA)-positive people, pigs and dust compared to farms with or without sows and finisher pigs. Number of farms (total 50) indicated in bars.

intensity of contact with pigs was another important determinant. In persons working with pigs on a regular basis, 29% (95% CI 20–38) carried MRSA whereas 12% (95% CI 3–29) of persons who did not work with pigs but entered the pighouse(s) at least once per week were positive. Only 2% (95% CI 0–6) of those who reported no contact with pigs were positive. On MRSA-positive farms, 49% of the people working with pigs harboured MRSA (95% CI 36–62).

At the farm level, the presence of sows related to a higher rate of MRSA in people. Several other factors were associated with MRSA positivity in humans, i.e. age, gender, sharing of towels, type of farm and number of pigs on farm. The presence of finisher pigs, number of new pig-batches recently received, and cleaning and disinfection measures in the pighouse did not influence MRSA rates.

No indication for other potential causes of MRSA was found. Four persons (on two farms) who were diagnosed with MRSA previously, proved negative at this sampling. Twelve family members worked in a hospital or nursing home but none of them carried MRSA. We did not find a relationship with recent hospitalization, team sports or presence of horses, cattle or other animals on the farm. MRSA appeared to be more frequent in people with skin problems ($P=0{\cdot}06$), but these were only reported by a small number ($n=17$).

MRSA per person on positive farms – multivariate analysis

On the 28 positive farms 139 people were sampled and the incidence of MRSA was 24%. Multivariate regression analysis of the results from the positive farms determined two significant risk factors for MRSA carriage out of 12 factors identified as potential risk factors in the univariate analysis. These were 'intensity of contact with pigs on the farm' and 'presence of sows and finishing pigs' (Table 2). The random cluster effect of 'farm' was not significant.

Genotyping MRSA

All MRSA isolates from human samples were Panton–Valentine leukocidin (PVL) negative. Seven different but related *spa*-types were found, most commonly t011 (45%), t567 (21%) and t108 (15%) which are all known NT-MRSA strains. Furthermore, two cases of t899 (known NT-MRSA) were found and also two t2330, one t2741 and one t588. The latter three *spa*-types were consequently submitted for PFGE analyses and also identified as NT-MRSA. In the pig and dust samples, t011 and t108 *spa*-types were most prevalent, as well as t567, t899 and t2330. Two *spa*-types, t588 and t2741, were found in people but not in pigs. Some *spa*-types found in pigs were not present in people.

On all 15 farms with MRSA-positive people, the *spa*-types in human samples matched with the types found in pigs on the same farm (30/33 persons). These were of *spa*-types t011 (eight farms), t108 (three farms), t567 (two farms), t2330 (one farm) and t899 (one farm). On 13/15 farms the type found in the pigs was the only *spa*-type found in people. However, on two farms persons were carrying another *spa*-type than found in the pigs. On one farm two co-workers harboured t108 and t2741 while the farmer and pigs carried t011; this farm also cared for other animals (sheep and horses). On the other farm, a household member yielded t588 while two other household members and the pigs had t108; on this farm a child had been previously diagnosed with MRSA.

Antimicrobial resistance patterns

All human MRSA isolates were resistant to tetracycline and all isolates were fully susceptible to vancomycin, teicoplanin, nitrofurantoin, fusidic acid and rifampin. Other antimicrobials showed variable resistance (Table 3).

The levels of resistance to ciprofloxacin, fosfomycin, tobramycin and cotrimoxazole were dependent on *spa*-types (χ^2, $P<0{\cdot}05$). Of the predominant *spa*-types, t011 showed higher levels of resistance to tobramycin and cotrimoxazole than the other *spa*-types, while t567 was more frequently resistant to

Table 1. *Human MRSA carriage of persons living on pig farms in relation to individual and farm-related characteristics on 50 pig farms in The Netherlands (left) and the on subgroup of 28 MRSA-positive farms (right), January–October 2007; univariate logistic regression analysis*

	Persons from all farms		Persons from MRSA-positive farms	
	Total	% MRSA+	Total	% MRSA+
Total	232	14·2	139	23·7
Individual factors				
Male	125	**20·8**	74	**35·1**
Female	107	**6·5**	65	**10·8**
Age (yr)				
0–18	80	**3·8**	48	**6·3**
19–65	138	**19·6**	84	**32·1**
66–86	14	**21·4**	7	**42·9**
Intensive contact with pigs	98	**28·6**	57	**49·1**
Minimal contact with pigs	25	12·0	14	**21·4**
No contact with pigs	109	**1·8**	68	**2·9**
Contact with cattle*	74	20·3	56	26·8
Skin problems reported*	17	29·4	11	45·5
Shared use of towels*	78	**21·8**	47	**36·2**
Farm-related factors				
MRSA-positive pigs on farm*	122	**26·2**	122	**26·2**
MRSA-positive dust on farm*	115	**25·2**	115	**25·2**
No MRSA-positive pigs on farm†	110	**0·9**	17	**5·9**
10–50 % of pool samples positive	30	**13·3**	30	**13·3**
60–100 % of pool samples positive	92	**30·4**	92	**30·4**
No MRSA-positive dust found†	117	**3·4**	24	16·7
20–60 % of dust samples positive	51	11·8	51	11·8
80–100 % of dust samples positive	64	**35·9**	64	**35·9**
Both sows and finishing pigs present	58	**25·9**	32	**46·9**
Only sows present	40	12·5	9	**55·6**
Only finishing pigs present	129	10·1	98	**13·3**
Only rearing pigs present	5	0·0	0	0·0
Small farm (<400 sows + finisher pigs)†	76	10·5	30	26·7
Medium farm (400–1000 pigs)	74	8·1	47	**12·8**
Large farm (>1000 pigs)	82	**23·2**	62	**30·6**
Personal hygiene in stable				
Low†	56	7·1	35	**11·4**
Intermediate	77	13·0	47	21·3
High	99	19·2	57	**33·3**

Percentages in bold are considered significant at $P<0{\cdot}05$ level.
* MRSA rate where factor present (shown) compared to factor absent (not shown).
† Divided into equal groups for optimal comparison.

fosfomycin and cotrimoxazole and less sensitive to ciprofloxacin.

Effect of short-term exposure of human and animal sample collectors

The research assistant who collected the human samples on 50 farms remained negative for MRSA throughout the research period, whereas 13/32 veterinary assistants collecting samples from pigs and stable dust had MRSA-positive samples on one or more occasions. On 13 farm visits (10 assistants), the collector was MRSA positive directly after sampling but negative by the next day. Two collectors were still positive the day after the visit; a sample from one of them 1 month later was negative, no repeat sample was available from the other. The *spa*-types identified from 11/12 collectors corresponded with the types

Table 2. *Risk factors for human MRSA identified by unilevel multivariate analysis, based on persons sampled at the 28 farms where pigs or dust samples were MRSA positive* (*n* = *139*)

	OR (95% CI)	*P* value multivariate
Intensive contact with pigs	39·9 (7·6–208·8)	**<0·0001**
Minimal contact with pigs	9·4 (1·2–73·5)	**0·03**
No contact with pigs	1·0	
Both sows and finishing pigs present	8·8 (2·6–29·9)	**0·001**
Only sows present	3·5 (0·8–16·6)	0·11
Only finishing pigs present	1·0	

OR, Odds ratio, CI, Confidence interval.
P values in bold are considered significant at $P < 0{\cdot}05$ level.

Table 3. *Susceptibility to antibiotics of human MRSA isolates* (*farm residents and collectors*), *by* spa-*type*

	Susceptible strains (%)				
	t011 (*n* = 28)	t108 (*n* = 16)	t567 (*n* = 8)	Other (*n* = 8)	Total (*n* = 60)
Nitrofurantoin	100	100	100	100	100
Vancomycin	100	100	100	100	100
Rifampicin	100	100	100	100	100
Linezolid	100	100	100	100	100
Mupirocin	100	100	100	100	100
Fusidic acid	100	94	87	100	97
Ciprofloxacin	79	87	25	75	74
Gentamicin	71	100	100	87	85
Tobramycin	68	100	100	87	67
Cotrimoxazole	36	81	25	50	48
Erythromycin	50	37	75	62	48
Clindamycin	50	37	75	62	48

found in the pighouses. One collector was already carrying MRSA before his first farm visit: he visited three farms and remained MRSA positive with *spa*-types not corresponding to the MRSA types on these farms.

DISCUSSION

This study shows that 30% of farms have MRSA-positive farm residents and 56% have MRSA present in pighouses. In all farm residents the incidence of MRSA was 14% and this doubled (29%) in persons working with pigs. The farms investigated were a representative sample of The Netherlands by region and type of farm, although the response rate (47%) may have caused a bias for farms cooperating because of previous knowledge on MRSA (and an interest in participating) or no knowledge (and no fear of participating). Nasal sampling is known to identify the majority of MRSA carriers [27]. Visits were spaced and samples carefully handled to avoid cross-contamination from one farm to another.

The transmission route of MRSA is probably from pigs to people. People were found to be positive only on farms with MRSA in pigs or dust, increasingly so with higher positivity rates in pigs. *Spa*-typing showed that 91% of people were colonized with similar strain(s) as the pigs on their farm. All MRSA strains were of NT-MRSA type and closely related. Most strains were of *spa*-types identified previously as NT-MRSA and of the animal-related MLST type ST398 [11, 28] and additional PFGE typing confirmed that all other *spa*-types were NT-MRSA. Two *spa*-types, t588 and t2741, were recovered from people but not pigs; more *spa*-types were found in pigs than in human samples.

Intensive and repeated exposure to positive pigs appears to be an important factor in MRSA colonization. MRSA positivity was common in persons working with pigs and also persons with less intensive but regular contact (weekly) with the animals. The MRSA transmission from pigs to humans was higher on farms with sows than on finisher farms, while MRSA in pigs circulated more frequently on finisher farms. Management of (breeding) sows (with regular deliveries, care of piglets) requires closer contact with pigs, especially with piglets, as well as longer working hours in the pighouse than management of finisher pigs and this may lead to a higher rate of MRSA transmission from pigs to humans. The high prevalence of NT-MRSA in dust samples from pighouses implies that MRSA could be spread by inanimate material as well, as has been postulated for hospital ICUs [29, 30].

The persistence of MRSA carriage is not determined in our point-prevalence survey. Assuming a continuous exposure, farm residents can be expected to remain MRSA positive, however, distinguishing persistent carriers would need repeated sampling [31, 32]. The results from repeated samples of our research teams imply that the risk of acquiring MRSA during short farm visits is limited to transient carriage, even when exposure to pigs is intense. It would be interesting to follow NT-MRSA carriage of pig farmers when they are no longer in contact with pigs (i.e. when on holiday or retiring).

The low MRSA rate (2%) in persons with no contact with pigs suggests a low level of human-to-human transmission of NT-MRSA. Hospital screening activities after detection of a MRSA carrier showed that animal-related ST398 MRSA led to fewer secondary cases (three from 24 index patients) than other MRSA genotypes (62 cases from 56 index patients [32]).

Impact of NT-MRSA in Dutch health-care system

The 14% MRSA carriage of pig-farm residents is much higher than the 0·12% prevalence in the open population and at primary care (studied in 2005–2006 [33]) and 0·03% at routine hospital admission (data from 1999–2000 [4]). Countrywide, there are about 9700 pig farms (National database CBS, 2005). Extrapolation of our results indicates that nearly 6000 MRSA carriers (range 4000–9500) may be expected in pig-farm residents. This justifies the hospital strategy to screen people having contact with pigs in Dutch hospitals since 2006 and agrees with a high proportion of NT-MRSA cases in the national MRSA surveillance database. Moreover, a recent report from hospitals showed that 30% of index patients carried animal-related MRSA [34].

The increase in MRSA-positive patients, identified by hospital screening, attributable to farm-animal MRSA will cause increased costs due to care in isolation, longer stay in hospital, specific diagnostics and medication [14]. However, the clinical relevance for the people concerned still needs further investigation. Transmissibility from human to human appears to be low and few symptomatic cases of NT-MRSA infections have been found [35].

Use of antimicrobials in pig farming and consequences for MRSA in humans

Antimicrobial selection pressure in general is one of the probable factors that have facilitated the emergence and spread of veterinary MRSA [28]. Antimicrobial consumption in pig farming in The Netherlands is substantial compared to other livestock farming [36]. Tetracyclines and trimethoprim–sulfonamide (trimsulfa) are most widely used; we found levels of resistance to these antibiotics of 100% and 52%, respectively. In other recent Dutch studies resistance to trimethoprim–sulfamethoxazole was not or hardly ever found in MRSA in pigs and humans [11, 12], hence this resistance might be currently emerging. The pattern of resistance of the MRSA samples in our study was otherwise comparable to that in other recent studies [11–13].

Implications for health care and future research

Since 2007, adjusted guidelines for hospitals in The Netherlands require screening for MRSA and care in isolation for all people professionally in contact with live pigs or veal calves. Our results, however, have shown that the frequency and intensity of contact with pigs, whether professional or not, are a determinant for MRSA risk and hence the terminology 'contact' should be refined further. The persistency of NT-MRSA carriage in humans after different types of exposure should also be studied further.

NT-MRSA appears to be frequently transmitted from pigs to people, but less so from person to person (the present study [15, 33]). The need for strict management of patients (isolated care) might be reviewed if human-to-human transmission of NT-MRSA is indeed as limited as shown.

Although the antimicrobial resistance pattern found here has no consequences yet for current treatment options of MRSA, further spread of NT-MRSA and selection of resistant strains by the high use of antimicrobials in pig farming, may impede the usefulness of antimicrobials in the future or necessitate differential treatment of MRSA and NT-MRSA (and thus T/NT-typing before treatment). The use of antimicrobials in pig farming should be studied in relation to MRSA prevalence and possible alternative treatment strategies investigated.

We found no association between personal hygiene and MRSA carriage, possibly because personal hygiene at the level that we investigated was not an important factor in pig-to-human MRSA transmission, and transmission of MRSA between humans did not seem to play an important role. Other factors, such as intensity of contact with animals and *actual use* of cleaning and protection methods may be more important. Protection methods might need to be adjusted to the type of pigs and activities in the stables. The role of hygienic measures in transmission reduction also requires further study.

NT-MRSA will be no doubt studied more extensively in other countries in Europe in the future. The pig-farming sector involves a wide European network of farms and the accompanying meat industry. Pig farms in other countries are probably facing a similar problem as the Dutch farms, although this has as yet not been reported as extensively. Action is needed at a

European level to assess the situation and design appropriate measures to prevent further spread of NT-MRSA.

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DECLARATION OF INTEREST

None.

REFERENCES

1. **Jevons MP.** Celbenin-resistant staphylococci. *British Medical Journal* 1961; **1**: 124–125.
2. **Cosgrove SE, *et al.*** The impact of methicillin resistance in *Staphylococcus aureus* bacteremia on patient outcomes: mortality, length of stay, and hospital charges. *Infection Control and Hospital Epidemiology* 2005; **26**: 166–174.
3. **Kluytmans-Vandenbergh MF, Kluytmans JA.** Community-acquired methicillin-resistant *Staphylococcus aureus*: current perspectives. *Clinical Microbiology and Infection* 2006; **12** Suppl 1: 9–15.
4. **Wertheim HF, *et al.*** Low prevalence of methicillin-resistant *Staphylococcus aureus* at hospital admission in the Netherlands: the value of search and destroy and restrictive antibiotic use. *Journal of Hospital Infection* 2004; **56**: 321–325.
5. **EARSS.** Annual report 2006, EARSS, Bilthoven, The Netherlands, October 2007. (www.rivm.nl/earss).
6. **Voss A, *et al.*** Methicillin-resistant *Staphylococcus aureus* in pig farming. *Emerging Infectious Diseases* 2005; **11**: 1965–1966.
7. **Bens CPM, Voss A, Klaassen CHW.** Presence of a novel DNA methylation enzyme in methicillin-resistant *Staphylococcus aureus* isolates associated with pig farming leads to un-interpretable results in standard pulsed-field gel electrophoresis analysis. *Journal of Clinical Microbiology* 2006; **44**: 1875–1876.
8. **Huijsdens XW, *et al.*** Community-acquired MRSA and pig farming. *Annals of Clinical Microbiology and Antimicrobials* 2006; **5**: 26.
9. **Vandenbroucke Grauls CMJE, Beaujean DJMA.** Methicillin-resistant *Staphylococcus aureus* in pig breeders and cattle breeders [in Dutch]. *Nederlands Tijdschrift voor Geneeskunde* 2006; **150**: 1710–1712.
10. **Wulf M, *et al.*** Methicillin-resistant *Staphylococcus aureus* in veterinary doctors and students, the Netherlands. *Emerging Infectious Diseases* 2006; **12**: 1939–1941.
11. **van Duijkeren E, *et al.*** Methicillin-resistant *Staphylococcus aureus* in pigs with exudative epidermitis. *Emerging Infectious Diseases* 2007; **13**: 1408–1410.
12. **de Neeling AJ, *et al.*** High prevalence of methicillin resistant *Staphylococcus aureus* in pigs. *Veterinary Microbiology* 2007; **122**: 366–372.
13. **Van Loo I, *et al.*** Emergence of methicillin-resistant *Staphylococcus aureus* of animal origin in humans. *Emerging Infectious Diseases* 2007; **13**: 1834–1839.
14. **Mooij R, *et al.*** MRSA in calves [in Dutch]. *Infectieziekten Bulletin* 2007; **18**: 234–236.
15. **Van Rijen MM, Van Keulen PH, Kluytmans JA.** Increase in a Dutch hospital of methicillin-resistant *Staphylococcus aureus* related to animal farming. *Clinical Infectious Diseases* 2008; **46**: 261–263.
16. **Armand-Lefevre L, Ruimy R, Andremont A.** Clonal comparison of *Staphylococcus aureus* isolates from healthy pig farmers, human controls and pigs. *Emerging Infectious Diseases* 2005; **11**: 711–714.
17. **Witte W, *et al.*** Methicillin-resistant *Staphylococcus aureus* ST398 in humans and animals, Central Europe. *Emerging Infectious Diseases* 2007; **13**: 255–258.
18. **Guardabassi L, Stegger M, Skov R.** Retrospective detection of methicillin resistant and susceptible *Staphylococcus aureus* ST398 in Danish slaughter pigs. *Veterinary Microbiology* 2007; **122**: 384–386.
19. **Denis O, *et al.*** High prevalence of 'livestock-associated' methicillin-resistant *Staphylococcus aureus* ST398 in swine and pig farmers in Belgium. In: *Abstracts of the 18th European Congress of Clinical Microbiology and Infectious Diseases*, 19–22 April 2008. Abstract O508.
20. **Hanselman BA, *et al.*** Methicillin-resistant *Staphylococcus aureus* colonization in veterinary personnel. *Emerging Infectious Diseases* 2006; **12**: 1933–1938.
21. **Khanna T, *et al.*** Methicillin resistant *Staphylococcus aureus* colonization in pigs and pig farmers. *Veterinary Microbiology* 2008; **128**: 298–303.
22. **Martineau F, *et al.*** Species-specific and ubiquitous DNA-based assays for rapid identification of *Staphylococcus aureus*. *Journal of Clinical Microbiology* 1998; **36**: 618–623.
23. **de Neeling AJ, *et al.*** Resistance of staphylococci in The Netherlands: surveillance by an electronic network during 1989–1995. *Journal of Antimicrobial Chemotherapy* 1998; **41**: 93–101.
24. **Lina G, *et al.*** Involvement of Panton-Valentine leukocidin-producing *Staphylococcus aureus* in primary skin infections and pneumonia. *Clinical Infectious Diseases* 1999; **29**: 1128–1132.
25. **Harmsen D, *et al.*** Typing of methicillin-resistant *Staphylococcus aureus* in a university hospital setting by using novel software for *spa* repeat determination and

database management. *Journal of Clinical Microbiology* 2003; **41**: 5442–5448.

26. **Murchan S, *et al.*** Harmonization of pulsed-field gel electrophoresis protocols for epidemiological typing of strains of methicillin-resistant *Staphylococcus aureus*: a single approach developed by consensus in 10 European laboratories and its application for tracing the spread of related strains. *Journal of Clinical Microbiology* 2003; **41**: 1574–1585.
27. **Mertz D, *et al.*** Throat swabs are necessary to reliably detect carriers of *Staphylococcus aureus*. *Clinical Infectious Diseases* 2007; **45**: 475–477.
28. **van Duijkeren E, *et al.*** Transmission of methicillin-resistant *Staphylococcus aureus* strains between different kinds of pig farms. *Veterinary Microbiology* 2008; **126**: 383–389.
29. **Dansby W, *et al.*** Aerosolization of methicillin-resistant *Staphylococcus aureus* during an epidemic in a burn intensive care unit. *Journal of Burn Care and Research* 2008; **29**: 331–337.
30. **Wilson RD, Huang SJ, McLean AS.** The correlation between airborne methicillin-resistant *Staphylococcus aureus* with the presence of MRSA colonized patients in a general intensive care unit. *Anaesthesia and Intensive Care* 2004; **32**: 202–209.
31. **Kluytmans J, van Belkum A, Verbrugh H.** Nasal carriage of *Staphylococcus aureus*: epidemiology, underlying mechanisms, and associated risks. *Clinical Microbiology Reviews* 1997; **10**: 505–520.
32. **Nouwen JL, *et al.*** Predicting the *Staphylococcus aureus* nasal carrier state: derivation and validation of a 'culture rule'. *Clinical Infectious Diseases* 2004; **39**: 806–811.
33. **NETHMAP.** In: Degener JE, Groningen UMC, de Neeling AJ, eds. Consumption of antimicrobial agents and antimicrobial resistance among medically important bacteria in the Netherlands. Report 2007. RIVM Bilthoven.
34. **Wassenberg MWM, *et al.*** Reduced transmission of animal-related ST398 methicillin-resistant *Staphylococcus aureus* in Dutch hospitals. In: *Abstracts of the 18th European Congress of Clinical Microbiology and Infectious Diseases*, 19–22 April 2008. Abstract P1405.
35. **van Belkum A, *et al.*** On behalf of the Dutch working party on surveillance and research of MRSA. Methicillin-resistant and -susceptible *Staphylococcus aureus* sequence type 398 in pigs and humans. *Emerging Infectious Diseases* 2008; **14**: 479–483.
36. **MARAN.** In: Mevius DJ, van Pelt W, eds. Monitoring of antimicrobial resistance and antibiotic usage in animals in The Netherlands in 2005. VANTURES, the Veterinary Antibiotic Usage and Resistance Surveillance Working Group, CIDC Lelystad, The Netherlands. 2006.