Quantitative exposure to livestock-associated MRSA ST398 of pig slaughterhouse workers

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Abstract
Objectives: To quantify livestock-associated MRSA (LA-MRSA) exposure to workers in pig slaughterhouses and assess associated risk factors for carriage in slaughterhouse workers.
Methods: A cross-sectional study in three Dutch pig slaughterhouses was undertaken. Nasal swabs of 341 participants, surface wipes, air, and glove samples were screened for presence of MRSA. MRSA was quantitatively determined on gloves and in air samples by culturing and real-time PCR.
Results: 3.2% of the participants were defined as nasal MRSA carrier. MRSA positive workers were predominantly found at the start of the slaughter process. Major risk factors for carriage were working in the lairage and working in the scalding and dehairing area. Most nasal isolates (73%) belonged to the LA-MRSA clone ST398. MRSA ST398 positive environmental samples were found throughout the slaughter process. A clear decrease was seen along the slaughter line in the number of MRSA positive samples and MRSA colony count per sample.
Conclusions: This study showed that working in the lairage area and scalding and dehauling area were the major risk factors for MRSA carriage in pig slaughterhouse workers, while the overall prevalence is low. Occupational exposure to MRSA decreased along the slaughter line and paralleled the risk of carriage. These results can be used to model occupational risk of MRSA carriage in related occupations with meat contact, such as butchers and cooks, which likely will also be very low.

Introduction
Livestock-associated Methicillin-resistant Staphylococcus aureus (LA-MRSA) has been increasingly prevalent in veal calves, pigs, and veal and pig farmers over the last decade. MRSA prevalence estimates in persons working or living on livestock farms in the Netherlands go up to 33% (1-3). However, MRSA prevalence was much lower in family members living on the farm (8%) and in veterinarians (12.5%) (2, 4). Human-to-human LA-MRSA transmission is still considered low compared to non-livestock-associated strains (5).
Most LA-MRSA studies aimed at people working or living on farms, but the question was raised whether slaughterhouse personnel also have increased LA-MRSA carriage risk, as they too come into contact with living animals, dead animals and meat products. Furthermore, it is unclear what the MRSA carriage risk is for other occupations with meat contact, such as butchers and cooks. Recent studies showed LA-MRSA prevalence to be around 5% in poultry and pig slaughterhouse personnel, and contact with live animals was established as the major risk factor for MRSA carriage (6, 7). Quantitative data on MRSA exposure from animals and their derived meat products to slaughterhouse workers however, are still lacking.
The study presented here quantifies MRSA load in air and on hands as an exposure estimate of workers in pig slaughterhouses, and assesses MRSA carriage risk factors. Results will be discussed in the light of implications for occupational health and consumer risks. The results can be used to model the occupational risk for other occupations with meat contact, such as butchers and cooks.
Material and Methods
A cross-sectional study was undertaken in three Dutch pig slaughterhouses in 2009 and 2010. Nasal swabs were collected from 341 workers and analysed to determine MRSA carriage by culturing of MRSA (standard procedure as described in [8]). MRSA was confirmed with a mecA specific PCR (9, 10). The genetic diversity of non-ST398 MRSA colonies cultured from nasal swabs was determined with MLST and spa-typing (11, 12). Furthermore, MRSA exposure in the air was measured by means of pumps taking active air samples, and by analysing the filters according to the procedures described above. Gloves of the workers were collected and presence of MRSA was quantitatively determined by culturing of MRSA, using a standard dilution curve, and by qPCR targeting ST398, SCCmec and mecA (9, 10, 13).

Questionnaires were used to collect data on potential risk factors and confounders: gender, age, smoking, country of birth, working area within slaughterhouse, contact with livestock outside of the slaughterhouse and recent antibiotics use. The data were analysed by means of logistic regression (SAS version 9.2).

Results
Eleven participants (3.2%) tested positive for nasal MRSA carriage. Most nasal isolates (73%) belonged to the LA-MRSA clone ST398. MRSA positive workers were predominantly found at the start of the slaughter process, i.e. in the lairage area, scalding and dehairing area, or evisceration area. Not surprisingly, major risk factors for carriage were working in the lairage area and scalding and dehairing area (OR: 5.6 and 4.1, respectively).

LA-MRSA positive glove samples were found throughout the slaughter process, with the highest levels at the front end. A clear decrease was seen along the slaughter line in the number of MRSA positive samples and MRSA colony count per air or glove sample, either by culturing or by qPCR calculated to estimated equivalent CFUs. Relatively twice as many culture positive gloves were found in the lairage and scalding and dehairing areas, when compared to the evisceration and cutting area. No culture positive gloves were found in the processing, packing and dispatch area. Quantitative PCR resulted in positive glove samples in the lairage, scalding and dehairing, evisceration, cutting, and processing area, with a clear decrease in average CFU's per sample along the processing line. Culture and qPCR positive MRSA air samples were found, but at low concentrations and only in the lairage area, and scalding and dehairing area.

Discussion
The study described here, confirmed that slaughterhouse workers were at low occupational risk of becoming MRSA carriers. The main risk factor was working in the lairage or scalding and dehairing area, in other areas exposure was found to be lower, both in air and glove samples. The overall carriage risk is considerably lower in slaughterhouse workers than in pig and veal farmers or veterinarians.

Research has shown that LA-MRSA prevalence in veal farmers decreased significantly after considerably reduced contact with veal calves, e.g. during the holiday or the empty-barn period between production rounds (14). Slaughterhouse workers may also be intermittent carrier. Therefore, they may, for example, loose the bacteria over the weekend as, so far, the bacteria seem to prefer the animal host over human hosts. Future studies should aim at establishing the duration of MRSA carriage in slaughterhouse workers.

General hygiene measures may help reducing risk of colonization or infection, such as regularly changing of gloves, and washing and disinfection of hands (all these facilities are provided in slaughterhouses). In addition, face masks could be properly worn to prevent inhaling LA-MRSA, especially in the lairage and scalding and dehairing areas. These hygiene measures could also be applied in other occupations with frequent contact with raw meat, such as butchers and cooks.

Previous studies have shown that MRSA could be found in meat samples, albeit in low concentrations and prevalence (15-17). It should be noted here, that the study by Pu et al. (2009) did not find zoonotic clones of MRSA, but the common hospital-acquired clone USA100 and the common community-acquired clone USA300. The study by De Boer et al. (2009) also found a significant part of the isolated MRSA strains from meat samples were non-ST398 strains. Of the strains isolated from gloves and air samples in the current study, 95% and 100% belonged to ST398, respectively. It remains unclear what the source is of the non-ST398 strains, but other sources than the animals or from outside the slaughterhouse are likely.

Although in the study described here the meat itself was not sampled, and therefore no conclusions can be drawn concerning meat products, qPCR positive glove samples were found in the cutting and processing areas. However, no positive samples were found in the packing or dispatching areas. Again, the number of positive samples was very low, as was the concentration of MRSA found in these samples. The supposed risk for occupations with contact with raw meat and even
consumers, is likely very low if they adhere to the standard (kitchen) hygiene protocols concerning raw meat products, and if they cook their meat properly (18). The data provided with this study will be used to model this risk.

Conclusion
This study showed that working in the lairage, scalding and dehairing area were the major risk factors for MRSA carriage in pig slaughterhouse workers, with a low overall prevalence. Occupational exposure to MRSA in air and on hands decreases along the slaughter line and parallels the risk of carriage. The results can be used to model occupational risk of MRSA carriage for other occupations with contact with raw meat.

References

