Acquisition of high-level mupirocin resistance in CoNS following nasal decolonization with mupirocin

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Objectives: The association between mupirocin use and plasmid-based high-level resistance development mediated through \( \text{mupA} \) in CoNS has not been quantified. We determined acquisition of mupirocin resistance in \( \text{Staphylococcus aureus} \) and CoNS in surgery patients treated peri-operatively with mupirocin.

Patients and methods: Patients admitted for surgery were treated with nasal mupirocin ointment and chlorhexidine soap for 5 days, irrespective of \( \text{S. aureus} \) carrier status. Nasal swabs were obtained before decolonization (T1) and 4 days after surgery (T2) and were inoculated onto agars containing 8 mg/L mupirocin. Staphylococci were identified by MALDI-TOF MS and mupirocin resistance was confirmed by Etest.

Results: Among 1578 surgical patients, 936 (59%) had nasal swabs obtained at T1 and T2; 192 (21%) patients carried mupirocin-resistant CoNS at T1 and 406 (43%) at T2 (\( P, 0.001 \)). Of 744 patients not colonized at T1, 277 acquired resistance (37%), corresponding to an acquisition rate of 7.4/100 patient days at risk. In all, 588 (97%) of 607 mupirocin-resistant CoNS had an MIC \( \geq 256 \text{ mg/L} \) (high level) and 381 of 383 (99.5%) were \( \text{mupA} \) positive. No acquisition of mupirocin resistance was observed in \( \text{S. aureus} \).

Conclusions: Acquisition of mupirocin resistance following decolonization was widespread in CoNS and absent in \( \text{S. aureus} \). As almost all isolates harboured the \( \text{mupA} \) gene, monitoring resistance development in \( \text{S. aureus} \) when decolonization strategies containing mupirocin are used is recommended.

Keywords: \textit{Staphylococcus aureus}, peri-operative decolonization, coagulase-negative staphylococci

Introduction

Mupirocin is a topical antibiotic and the cornerstone of decolonization regimens for MSSA and MRSA in patients and healthcare personnel. Peri-operative eradication of \textit{Staphylococcus aureus} nasal carriage, with mupirocin and chlorhexidine body washings, reduces the incidence of post-operative \textit{S. aureus} infections by 58%.1 However, the rapid identification of \( \text{S. aureus} \) carriers and immediate application of treatment is logistically challenging and costly. Therefore, universal peri-operative decolonization, irrespective of \( \text{S. aureus} \) carrier status, would be more cost-effective.2 Yet, extensive use of mupirocin, as in universal decolonization, may facilitate emergence of mupirocin resistance in \( \text{S. aureus} \) and CoNS, but this risk has not been quantified. We therefore investigated the effects of universal decolonization with topical mupirocin and chlorhexidine body washings on resistance in CoNS and \( \text{S. aureus} \).

Patients and methods

Setting and patient population

This study was performed at a tertiary teaching hospital in Utrecht, The Netherlands. A universal decolonization strategy was implemented on three surgical wards: a cardiothoracic surgery ward, an orthopaedic surgery ward and a neurosurgical ward for all patients undergoing surgery with an expected stay of \( \geq 4 \) days, using mupirocin nasal ointment three times daily for 5 days and chlorhexidine body washings once daily for 5 days, starting decolonization on the day of admission. On the three surgical wards, nasal swabs for detection of mupirocin-resistant staphylococci were taken from all consecutive patients treated with mupirocin, between June 2012 and June 2013, before the start of decolonization treatment (T1) and 4 days after surgery (1 day after completing decolonization treatment) (T2). A nasal swab was also obtained if patients were discharged or transferred to another hospital before completing the 5 days of decolonization therapy. The monitoring of resistance on...
Patients colonized with mupirocin-resistant CoNS at T1, 129 (67%) were still colonized at T2 and 63 (33%) no longer had detectable colonization at T2. All 13 patients colonized at admittance with an intermediate resistant CoNS (MIC range 8–256 mg/L) lost colonization by intermediate isolates at T2. Six of these 13 patients acquired a high-level resistant strain.

None of the 17 geriatric patients was colonized with mupirocin-resistant CoNS at T1 or T2.

Overall, 607 mupirocin-resistant CoNS were identified in 469 patients, of which 588 (97%) had high-level resistance (MIC $\geq$512 mg/L). Almost all were S. epidermidis (568/607, 94%). Mupirocin-resistant S. aureus were not detected in 939 patients with swabs taken at T1 and T2. One patient carried a mupirocin-resistant S. aureus (MIC 512 mg/L) at admission, with no swab taken at T2. Antibiotic susceptibility patterns besides mupirocin were determined in 100 randomly selected CoNS, revealing resistance to oxacillin in 69%, aminoglycosides (either tobramycin or gentamicin resistance) in 61%, clindamycin in 61%, ciprofloxacin in 62%, trimethoprim/sulfamethoxazole in 51% and rifampicin in 8%.

A PCR for the detection of mupA was performed on 383/607 (63%) strains. mupA was detected in 381 of 383 strains (99.5%) of high-level mupirocin-resistant CoNS and in four of 14 (29%) CoNS with intermediate resistance, with MICs ranging from 8 to 128 mg/L in mupA-positive strains. MLVA typing of 75 isolated high-level resistant S. epidermidis strains from the three surgery wards yielded 15 different genotypes, without a major dominant clone or evidence of clonal spread among those who acquired mupirocin-resistant S. epidermidis (Table S1).

### Results

During the study period, 1578 surgical and 22 geriatric patients were screened on admittance, from whom in 936 (59%) surgical and 17 (77%) geriatric patients nasal swabs were obtained both at T1 and T2. Subsequent analysis was performed on the 936 surgical patients (930 unique patients and 6 readmissions) and 17 control patients with swabs taken at both T1 and T2. In surgical patients, mupirocin-resistant CoNS were detected at T1 and T2 in 192 (21%) and 406 (43%) patients, respectively ($P<0.001$) (Table 1). Of the 744 patients not colonized at T1 and thus at risk of acquisition of mupirocin-resistant staphylococci, 277 (37%) acquired colonization at T2. This corresponds to an acquisition rate of 7.4/100 patient days at risk. Of the 192 patients colonized with mupirocin-resistant CoNS at T1, 129 (67%) were still colonized at T2 and 63 (33%) no longer had detectable colonization at T2. All 13 patients colonized at admittance with an intermediate resistant CoNS (MIC range 8–256 mg/L) lost colonization by intermediate isolates at T2. Six of these 13 patients acquired a high-level resistant strain.

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### Discussion

In this prospective cohort study, the prevalence of high-level resistance against mupirocin among CoNS was 21% before surgery and this increased to 43% after topical treatment with mupirocin and chlorhexidine body washing. Mupirocin-resistant S. aureus was only detected in one of 1578 patients at admission and no acquisition of mupirocin resistance was observed.

In this study, we screened for mupirocin-resistant staphylococci with an MIC $\geq$16 mg/L, thereby ignoring intermediate or low-level resistant isolates with MICs <16 mg/L. Low-level mupirocin resistance in S. aureus is mediated through mutations in the native iieRS, whereas the plasmid-based mupA gene encodes high-level resistance. We focused on mupA-positive staphylococci because of the potential of this gene to spread between species in vitro and in vivo. Because 99.5% of the high-level resistant isolates harboured mupA, we did not look for the presence of mupB, an alternative gene encoding mupirocin resistance. In addition to mupA, the qacA/B genes encoding resistance to antibiotics could potentially provide an additional source for cross-transmission between staphylococcal species and are commonly found in conjunction with low-level mupirocin resistance.

Clonal dissemination of mupirocin-resistant S. aureus and CoNS in hospital settings has been described. Based on our MLVA typing, we were able to exclude the possibility that cross-transmission fuelled the acquisition during treatment. mupA has been detected in all major dominant MRSA clones, suggesting
that interclonal transfer of mupA could contribute to the spread of mupirocin resistance.\textsuperscript{9}

This study has several limitations. First, no broth enrichment was used, potentially underestimating the prevalence of \textit{S. aureus}. Second, only a short period of follow-up was available for all patients, potentially underestimating the transfer rate of mupirocin resistance from CoNS to \textit{S. aureus}. However, the universal decolonization strategy had already been implemented for almost a year on the cardiothoracic ward and only one mupirocin-resistant \textit{S. aureus} was found. Third, no typing was performed on strains from patients colonized both at T1 and T2. Finally, as mupirocin-susceptible \textit{S. aureus} and CoNS were not collected, we could not differentiate whether acquisition of mupirocin-resistant CoNS during decolonization resulted from resistance development of the colonizing strain, from selection of a pre-existent mupirocin-resistant strain under antibiotic pressure or from exogenous acquisition. We could also not assess the percentage of patients with nasal co-carriage of mupirocin-resistant CoNS and mupirocin-susceptible \textit{S. aureus} at admission.

Transfer of mupA between \textit{S. epidermidis} and \textit{S. aureus} has been described during mupirocin prophylaxis \textit{in vitro} and \textit{in vivo}.\textsuperscript{4} However, no mupirocin-resistant \textit{S. aureus} were found at T2 during the study period, suggesting that horizontal transmission of the mupA gene from CoNS to \textit{S. aureus} did not occur in $\geq$38 patients (assuming 20% of the patients are \textit{S. aureus} carriers) carrying resistant CoNS at the start of treatment and with follow-up cultures obtained. Furthermore, no acquisition of mupirocin resistance in \textit{S. aureus} was observed, which was in stark contrast to 37% of the patients acquiring mupirocin resistance in CoNS. The differences in acquisition rates between \textit{S. aureus} and \textit{S. epidermidis} are poorly understood and should be studied in future work.

Based on its beneficial cost-effectiveness profile, we recommend universal decolonization with mupirocin nasal ointment and chlorhexidine body washing. However, the pre-existing high prevalence and rapid increase of plasmid-based high-level mupirocin resistance in CoNS during treatment warrant careful monitoring of mupirocin resistance development. In our study, horizontal gene transfer of mupA to \textit{S. aureus} was not demonstrated in 936 patients during the 5 days of follow-up. Future studies determining the persistence of carriage with high-level mupirocin-resistant CoNS and quantifying the horizontal transfer rate in patients with longer follow-up are needed for more accurate assessment of the ecological safety of universal decolonization with mupirocin in surgical and critically ill patients.

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### Transparency declarations

None to declare.

### Supplementary data

Supplementary data, including Table S1, are available as Supplementary data at JAC Online (http://jac.oxfordjournals.org/).

### References