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*Article*

# Methicillin-Resistant *Staphylococcus aureus* (MRSA) Contamination in Bedside Surfaces of a Hospital Ward and the Potential Effectiveness of Enhanced Disinfection with an Antimicrobial Polymer Surfactant

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**Abstract:** The aim in this study was to assess the effectiveness of a quaternary ammonium chloride (QAC) surfactant in reducing surface staphylococcal contamination in a routinely operating medical ward occupied by patients who had tested positive for methicillin-resistant *Staphylococcus aureus* (MRSA). The QAC being tested is an antibacterial film that is sprayed onto a surface and can remain active for up to 8 h. A field experimental study was designed with the QAC plus daily hypochlorite cleaning as the experimental group and hypochlorite cleaning alone as the control group. The method of swabbing on moistened surfaces was used for sampling. It was found that 83% and 77% of the bedside surfaces of MRSA-positive and MRSA-negative patients respectively were contaminated with staphylococci at 08:00 hours, and that the staphylococcal concentrations increased by 80% at 1200 h over a 4-hour period with routine ward and clinical activities. Irrespective of the MRSA status of the patients, high-touch surfaces around the bed-units within the studied medical ward were heavily contaminated (ranged 1 to 276 cfu/cm$^2$ amongst the sites with

positive culture) with staphylococcal bacteria including MRSA, despite the implementation of daily hypochlorite wiping. However, the contamination rate dropped significantly from 78% to 11% after the application of the QAC polymer. In the experimental group, the mean staphylococcal concentration of bedside surfaces was significantly ($p < 0.0001$) reduced from $4.4 \pm 8.7$ cfu/cm$^2$ at 08:00 hours to $0.07 \pm 0.26$ cfu/cm$^2$ at 12:00 hours by the QAC polymer. The results of this study support the view that, in addition to hypochlorite wiping, the tested QAC surfactant is a potential environmental decontamination strategy for preventing the transmission of clinically important pathogens in medical wards.

**Keywords:** MRSA; staphylococcal infections; surface contamination; environmental contamination; quaternary ammonium chloride; JUC; antimicrobial surfactant

## 1. Introduction

Microorganisms on hospital surfaces can be transmitted to the hands of healthcare workers, patients, and visitors, resulting in cross-infections and epidemics. Despite the implementation of routine cleaning and precautionary measures in most hospitals, effective environmental decontamination methods are still in demand. In recent decades, numerous polymeric surfactant products have been shown to have excellent antimicrobial properties against surface contamination, but none have been tested on hospital surfaces [1,2]. JUC spray is a nano-scale technology formulated with cationic organosilicon quaternary ammonium chloride (OrganoSiQAC) as a major ingredient that is currently being marketed as an FDA-approved invisible hydrogel antimicrobial dressing for wound care. According to a local case report, the JUC spray has also been demonstrated to be effective in managing MRSA-associated skin abscesses [3]. Two recent trials [4,5] have demonstrated a reduction in bacterial burdens from using JUC polymer on critical medical surfaces. These included urinary catheters, where the associated incidence of infection was significantly reduced [5]. The manufacturer of JUC claims that this antimicrobial film stays on animate and non-animate surfaces for up to eight hours [1]. Our research team was therefore particularly interested in investigating the long-acting surfactant capacity of JUC polymer on hospital surfaces and its potential application as an effective decontamination aid.

In hospitals, surfaces with which patients have close contact or that are highly accessible to patients are more likely to become contaminated. Environmental MRSA contamination has been extensively reported in different areas of a hospital, including in intensive care units [6–9], burn units [10], isolation rooms [11,12], and general wards [13]. In acute hospital wards, MRSA can be recovered from 1%–27% of surfaces in MRSA-positive patient rooms [14]. However, the incidence of MRSA contamination varies among different hospital ward surfaces, as contamination is influenced by various factors such as the condition of the patient, the ward setting, crowding, and even the sampling method [15,16]. It has been well documented that high-touch surfaces are major reservoirs for MRSA in hospital environments. Of all hospital surfaces, bedside rails in wards occupied by MRSA patients have been identified as the site most frequently contaminated with MRSA [14]. Other frequently contaminated surfaces include bed cranks, overbed tables, bed linens, bedside lockers, bedside trays, pressure cuffs, intravenous pumps, curtains, door handles, keyboards, and floors [6,9,11,13].

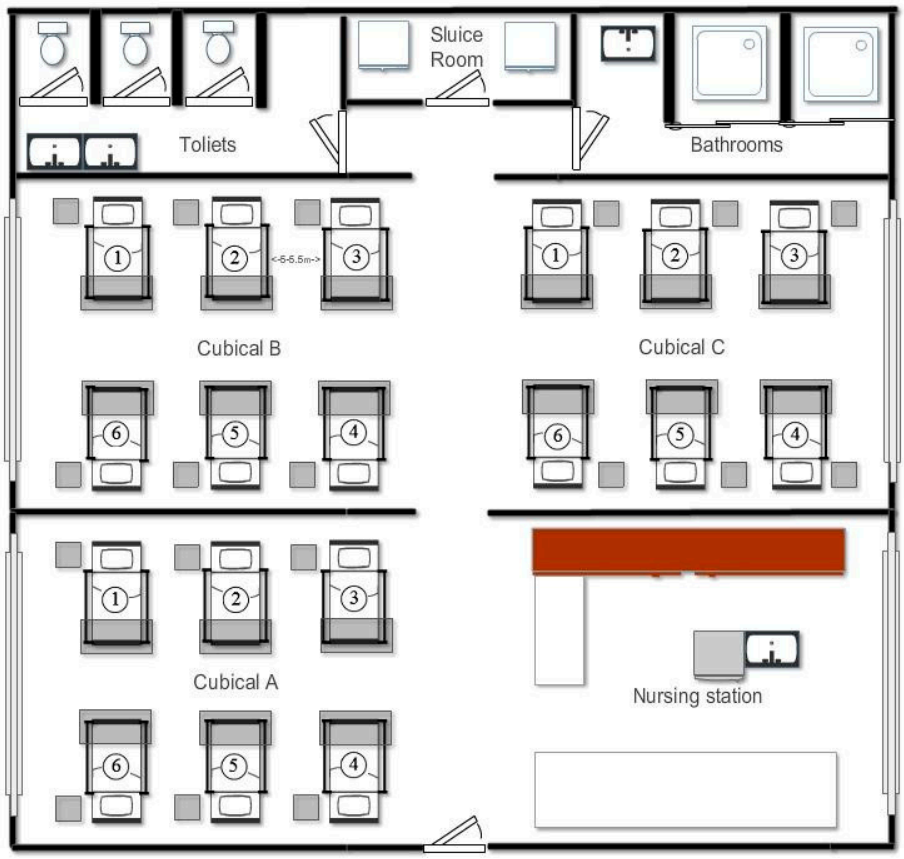
The prevalence of hospital-associated MRSA (HA-MRSA) infection varies geographically. Hong Kong is one of the high-prevalence areas in Asia [17]. According to the Asian Network for Surveillance of Resistant Pathogens (ANSORP) study, 57% of all inpatient isolates of *S. aureus* from Hong Kong hospitals were shown to be methicillin resistant [18]. In Hong Kong, healthcare services are provided by the government-supported Hospital Authority. In public hospitals, general wards are typically arranged in the setting of six beds per cubicle. Unlike in many other countries, known cases of MRSA colonization or infection at admission would not be isolated or assigned to single rooms. Rather, such patients would occupy beds at the far end of a ward. Since the surfaces of bedside environments are not considered critical surfaces in terms of contact with mucosal membranes, such surfaces are cleaned with hypochlorite wipes once a day in accordance with the environmental infection control strategies of the U.S. Centers for Disease Control and Prevention (CDC) [15]. Protective barrier precautions, such as the use of gloves and masks, are taken when handling MRSA patients. In view of the high MRSA infection rate in Hong Kong and the approach to dealing with MRSA-positive patients in local medical wards, the primary aim of this study was to evaluate the decontamination effectiveness of JUC polymer on bedside surfaces in a routine-operating medical ward in addition to the daily cleaning hypochlorite wipe and protective barrier precautions. To the best of our knowledge, there has been no epidemiological study thus far on surface MRSA-related contamination in the general wards of Hong Kong hospitals. The degree to which highly accessible bedside surfaces within a medical ward are contaminated with MRSA and other staphylococcal bacteria was assessed before the JUC polymer was evaluated, particularly when the ward was occupied by an MRSA-positive patient.

## 2. Experimental Section

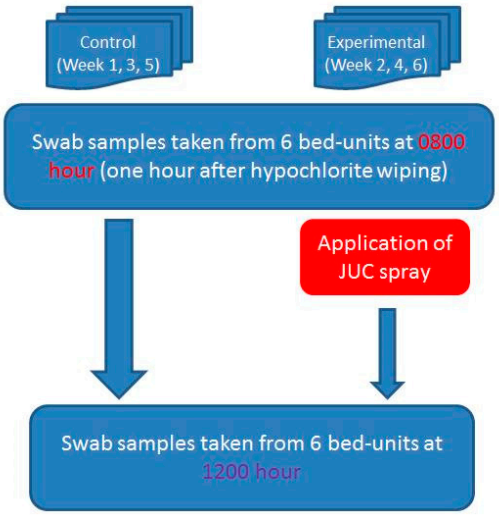
A field experimental study was designed to assess the degree of staphylococcal contamination on the bedside surfaces of a medical ward, and the effects of JUC polymer on reducing such contamination. Routine operations continued to be carried out in the ward to reflect the most natural environment for assessment.

### *2.1. Setting of the Study Hospital Ward*

The study hospital was a large teaching hospital in Hong Kong with a capacity over 1500 beds. The study ward was a male medical ward, consisting of a total of three cubicles each containing six beds, which were fully occupied on all of the days during the study period. The ward and cubicle setting is illustrated in Figure 1a. Cubicle A was designated for this study. Each cubicle was 53 by 60 square meters in area, and each bed-unit occupied 21 by 9 square meters with a bed-to-bed distance of 5 to 5.5 m. An indoor temperature of 24.5 to 25.5 degrees Celsius and humidity of 50 to 55 percent were maintained in the ward. The ward had 24-hour air conditioning with a proper ventilation system, and all of the windows were closed. Sampling to assess the contamination and experiments to evaluate the JUC polymer were carried out only on the days when the cubicle was occupied by one known patient with MRSA infection or colonization for not less than 24 h. In accordance with the usual practice, the MRSA-positive patients occupied bed number one of the cubicle (Figure 1a).




(**a**)




(**b**)

**Figure 1.** (**a**) Ward layout with three cubicles, and each cubicle consisting of six bed-units. Experiments and environmental sampling were conducted in Cubicle A, as it is separated from the other two cubicles and relatively remote from the sluice room, bathrooms, and toilets. On all sampling days the MRSA patients occupied Bed-unit 1 at the far end. (**b**) Experimental schedule for the study and control arms and to illustrate 6 weeks were required for the whole experiment because there was a week washing out period after each sampling day, in order to avoid the carry-over effects (if any) of the JUC spray.

### *2.2. Assessing Staphylococcal Contamination on Bedside Surfaces*

Four high-touch bedside surface sites (including the bedside table, the left-side handrail, the right-side handrail, and the overbed rolling table) were selected for the swab sampling. All of the selected sites had been identified as high-touch surfaces using a quantitative approach [19]. The sampling method was tested before the main study was commenced. The pilot results also indicated a remarkable degree of contamination (83% of the cultured surface specimens was found to be positive for staphylococci ranged in average 1 to 27 CFU/cm$^2$ per sampling site) at all of the selected sites. Using flocked or macrofoam swabs were found to be a reliable and efficient strategy for recovering bacteria from environmental surfaces [20]. In this study, the "swabbing on moistened surfaces" method was adopted using Puritan® sterile macrofoam tip environmental swab applicators for swab sampling. Swabbing was performed at three random spots of each sampling surface standardized to 10 square centimeters. Hence, a total of 12 swab specimens (three samples per site × four spots) were collected from each bed-unit. The swabbed specimens were inoculated into vials containing 1 mL of sterile water and transferred to Petrifilm™ Staph Express Count Plates (3M, St. Paul, MN, USA).

The degree of contamination in the bed-units occupied by MRSA-positive and MRSA-negative patients was compared. Swab specimens were collected from all bed-units within the cubicle at two time points at 08:00 hours and 12:00 hours, and this was repeated on six separate days over a 6-week period. The time of eight o'clock in the morning was chosen because that is not long after hypochlorite wiping has been done, at 07:00 hours, and most patients are awake following the minimal activities during the night shift within the ward. The hypochlorite solution was freshly prepared according to hospital guidelines by a duty healthcare worker, who also performed the wiping of the whole cubicle under the supervision of a duty nurse to ensure that all surfaces were cleaned. Hence, the 08:00 hours specimens were used as the baseline for comparisons between the MRSA-positive and MRSA-negative surfaces. Furthermore, the 12:00 hours specimens were used to assess natural changes in contamination over the four-hour interval during which routine clinical activities were carried out without disturbance. During the four-hour period before the lunch hours, visitors are generally not allowed to enter the ward, which minimizes disturbances other than those from patients and healthcare workers. However, healthcare professionals outside the ward cannot be prevented from entering the ward. Over the course of the 6-week study, a total of 864 swab specimens (three samples per site × four spots × six beds × two time points × six days) were collected from the designated site surfaces.

### *2.3. Experimental Design for Evaluating the Decontamination Effects of JUC Polymer*

In this experiment, the decontamination property of the JUC polymer was evaluated on the same designated sites of the bed-units used for the staphylococcal assessment in the same ward cubicle. The experimental arm tested the JUC spray plus the routine daily cleaning (*i.e.*, sanitization with bleaching water at 07:00 hours), while the control arm tested the routine daily cleaning alone. Regardless of the study arm, swab specimens were collected at 08:00 hours as the baseline and at 12:00 hours for assessing the change in the staphylococcal burden. The JUC dressing spray was gifted by the NMS Technologies Company (Nanjing, China). On the experimental days, the JUC spray was applied to the site surfaces immediately after the baseline specimens had been collected (see Figure 1b). The

following spraying method recommended by the manufacturer was followed: the target surface was evenly sprayed at a distance of 10 cm for 5 s to cover the spot area. No dangerous aerosol should be generated when spraying, since the JUC spray is 98% composed of water. The JUC spray was applied to all bed-units within the cubicle and the process was repeated three times over a 3-week period, for a total of 72 spots, *i.e.*, four sites per bed × six beds per cubicle × three experimental days. Since three random swabs were collected from each surface site, there were a total of 216 specimens per experiment or control arm. The swabbed specimens were inoculated into vials containing 1 mL of sterile water and transferred to Petrifilm™ Staph Express Count Plates (3M). To avoid a carry-over effect from the JUC spray, each experimental day was arranged to be at least 7 days apart from the next experimental day. The control arm was also conducted on three separate days. On each experimental and control day, one MRSA-positive patient occupied the bed-unit at the far end, *i.e.*, Bed-unit A1 in Figure 1. The ward staff and healthcare workers were blinded to the experiment in order to ensure that the routine ward activities were not influenced and disturbed. The demographic characteristics of the patients occupying the bed-units, including their MRSA status, are summarized in Table 1.

**Table 1.** Demographic characteristics of patients who occupied the bed-units in the experimental and control days.

| Parameters | | Experimental Group (n = 18) | Control Group (n = 18) | Paired *t*-Test |
|---|---|---|---|---|
| Mean Age ± SD (Range) | | 81.13 ± 7.17 (67–91) | 73.44 ± 16.26 (38–92) | 0.1041 |
| Total number of patients | | 18 | 18 | N.A. |
| Admitted from elderly homes | Yes | 11 (61%) | 10 (63%) | N.A. |
| (Number of patients) | No | 7 (39%) | 6 (37%) | N.A. |
| Positive culture for MRSA[#] | Wound swab | 1 | 1 | N.A. |
| (Number of patients) | Nasal swab | 0 | 1 | |
| | Sputum | 2 | 1 | N.A. |
| | None | 15 | 15 | N.A. |
| Mean hospitalization days ± SD (Range) | | 7.81 ± 4.35 (2–16) | 6.75 ± 2.95 (1–14) | 0.4395 |

Note: A total of 18 patients (with three MRSA-positive patients in three separate days) were involved in each arm of the study (experiment: JUC spray + standard cleaning verses control: standard cleaning). They were studied over three separate days (each 7 days apart). On each day, all six beds were fully occupied, with one MRSA carrier and five non-MRSA carriers within the ward cubicle.

### *2.4. Bacterial Culture and Identification*

Within one hour following the collecting of samples, all inoculated Staph Express Count Plates were sent to the microbiology laboratory of the study hospital for incubation and further analysis. All of the plates were incubated at 37 degrees Celsius for 24–48 h. Red-violet colonies were counted as positive for staphylococcal growth and expressed in Colony-forming Units per Centimeter Square (cfu/cm$^2$). Since each swab sample was collected from a surface area of 10 cm$^2$, the CFU/Petrifilm actually represented the growth of staphylococcal colonies from each swab specimen covering 10 cm$^2$ surface area. The minimum detection (countable) limit of the sampling technique was defined as 1 cfu/cm$^2$. Positive growth on the plates was followed up using the coagulase test for differentiating

coagulase-positive and coagulase-negative staphylococci (CNS). Subculture of the coagulase-positive staphylococci (red-violet colonies were randomly picked from Staph Express Count Plates) was performed in Mueller-Hinton agar supplemented with 4% w/v sodium chloride, and their sensitivity to oxacillin was assessed by using the disc diffusion test as recommended by the U.S. Centers for Disease Control and Prevention (http://www.cdc.gov/HAI/settings/lab/lab_mrsa.html). MRSA strains are resistant to all β-lactam antibiotics, but typically oxacillin resistant. Therefore, in this study, methicillin-resistant *Staphylococcus aureus* (MRSA) strains were identified and differentiated from the methicillin-sensitive *Staphylococcus aureus* (MSSA) strains. According to the technical bulletin and user instruction of the JUC spray dressing, the QAC solution will be solidified immediately after contacting the air after spraying onto any surface, and its bactericidial property is exerted by the physical electrostatic force generated between the positively-charged coating surface and the negatively-charged cell wall or membrane of the organisms. The coating adhered on the surface will then be water-proof and stay for up to 8 h. Such coating should not be removed from the sampling surface when performing the swab. Furthermore, physical bactericidal activity will be lost even if the coating is being released in solution, and therefore, neutralizer is not required for the specimen collection procedure from the bedside surface with the JUC coating and residual QAC is not a concern that could affect the microbiological results.

### *2.5. Ethical Considerations*

The experimental procedure was reviewed and approved by the ethics committee of the Hong Kong Polytechnic University as well as that of the study hospital. This study involved only the environmental surfaces around the bed-units inside a ward cubicle, where neither patients nor ward staff were involved. The JUC spray is an FDA-approved wound care product that causes no harm to humans. At admission, the MRSA status of the patients was confirmed according to the patients' history and to information provided by the hospital infections control team, and this information was not disclosed anywhere to anyone.

## 3. Results

At 08:00 hours, positive staphylococcal growth was recovered from 83% (five out of six) and 77% (23 out of 30) of the bedside surfaces occupied by MRSA-positive and MRSA-negative patients, respectively in the studied ward cubicle (Table 2). Irrespective of MRSA carrier status, both coagulase-positive (CPS) and coagulase-negative staphylococci (CNS) were recovered from all bed-units. Three random swab specimens were collected at 08:00 hours (an hour after hypochlorite cleaning) from each bedside surface of each bed-unit, thus a total of 108 swabs from each bedside surface site tested. Majority of the sampled bedside surfaces were negative for the bacterial growth (4.6%–11.1% positive for CNS; 2.8%–9.3% positive for MSSA; and 7.4%–17.6% positive for MRSA as summarized in Table 3). Among those surfaces with positive growth, the colony numbers recovered for CNS, MSSA, and MRSA were ranged 1–16, 1–193, and 1–276 cfu/cm$^2$, respectively. Irrespective of the swabbing site, CNS, MSSA, and MRSA were recovered from 44%, 28%, and 56% of all bed-units sampled. The mean CNS concentrations in the bed-units of the MRSA-positive and MRSA-negative patients were 1.9 cfu/cm$^2$ and 1.6 cfu/cm$^2$, respectively. From the bed-units of the MRSA-positive

patients, the ratio of CPS and CNS was found to be 2:4, and all CPS isolates were revealed to be oxacillin resistant with an overall burden of 3.9 cfu/cm$^2$. On the other hand, from the MRSA-negative bed-units, the CPS isolates were a mixture of MSSA and MRSA. Half of the CPS isolates were revealed to be oxacillin resistant with a mean concentration of 7.9 cfu/cm$^2$, which was significantly ($p < 0.05$) heavier than the amount recovered from the MRSA-positive bed-units. Overall, the staphylococcal distribution among CNS, MSSA, and MRSA was in the ratio of 2:6:8. Reference to the layout of the testing ward cubicle (Figure 1a), heavier growths were found to be at beds next to the MRSA-positive bed (*i.e.*, beds 2 and 6). Bed 4 was also observed to have relatively heavy growth. However, none of the beds had growth that was statistically different from that of other beds (data is not shown). Among all control bed-units received only hypochlorite cleaning, the mean staphylococcal contamination increased significantly ($p < 0.01$) by 80% from 08:00 to 12:00 hours (Figure 2a).

**Table 2.** A comparison of the concentrations and types of staphylococcal bacteria recovered from the bedside surfaces of bed-units occupied by MRSA-positive and MRSA-negative patients at 08:00 hours.

| Types of Staphylococci and Sampling Sites | Mean cfu/cm$^2$ ± Standard Error of Mean (SEM) | | |
|---|---|---|---|
| | MRSA-Positive Bed-Units (n = 6) | MRSA-Negative Bed-Units (n = 30) | *p* Value |
| CNS | | | 0.1953 |
| Bedside table | N.D. | 0.04 ± 0.03 | |
| Left-side handrail | 0.39 ± 0.33 | 0.12 ± 0.09 | |
| Right-side handrail | 1.94 ± 1.13 | 1.20 ± 1.13 | |
| Overbed rolling table | N.D. | 0.29 ± 0.25 | |
| MSSA | | | Undetermined |
| Bedside table | N.D. | 0.48 ± 0.39 | |
| Left-side handrail | N.D. | 1.94 ± 1.83 | |
| Right-side handrail | N.D. | 3.08 ± 2.17 | |
| Overbed rolling table | N.D. | 0.02 ± 0.01 | |
| MRSA | | | 0.0392 |
| Bedside table | N.D. | 0.68 ± 0.63 | |
| Left-side handrail | 0.72 ± 0.72 | 6.37 ± 4.03 | |
| Right-side handrail | 1.11 ± 0.98 | 0.57 ± 0.30 | |
| Overbed rolling table | 2.11 ± 1.41 | 0.28 ± 0.14 | |

Notes: A total of 36 bed-units were sampled in 6 separate days at 08:00 hours. The studied cubicle was fully occupied in all of those days, and only one MRSA-positive patient was included in each sampling day. The statistical difference in MSSA loadings between the MRSA-positive and MRSA-negative could not be determined, because the count for one comparison group was zero. N.D. = Non-detectable which means colony is absent in the petrifilm plate. The detection limit of the sampling technique was 1 cfu/Petrifilm (which is equivalent to cfu/cm$^2$). Some of the mean cfu/cm$^2$ values presented in this table were smaller than the detection limit as influenced by a large number of 0 (non-detectable) values (refer to Table 3 for the percentage of the positive culture at each sampling site).

**Table 3.** Number, percentage and the range of colony numbers of the three staphylococcal species recovered from the surface swabbing sites at 08:00 hours (an hour after the hypochlorite cleaning).

| Types of Staphylococci and Parameters | Site for Surface Swabbing (n = 108 for Each Site) | | | |
|---|---|---|---|---|
| | **Bedside Table** | **Left-Side Handrail** | **Right-Side Handrail** | **Overbed Rolling Table** |
| CNS | | | | |
| Number (%) of positive culture | 5 (4.6) | 6 (5.6) | 12 (11.1) | 7 (6.5) |
| Range of cfu/cm$^2$ among positive culture | 1–2 | 1–9 | 1–16 | 1–14 |
| MSSA | | | | |
| Number (%) of positive culture | 7 (6.5) | 10 (9.3) | 4 (3.7) | 3 (2.8) |
| Range of cfu/cm$^2$ among positive culture | 1–36 | 1–164 | 1–193 | 1–2 |
| MRSA | | | | |
| Number (%) of positive culture | 8 (7.4) | 19 (17.6) | 19 (17.6) | 12 (11.1) |
| Range of cfu/cm$^2$ among positive culture | 1–23 | 1–276 | 1–75 | 1–26 |

The same trend was observed in the bed-units of MRSA-positive and negative patients; however, the increase in staphylococcal contamination in the MRSA-negative beds (from 2.8 to 4.8 cfu/cm$^2$) was approximately four times that in the MRSA-positive bed-units (from 1.5 to 2.0 cfu/cm$^2$) (Figure 2b,c). The staphylococcal strain that increased after the four-hour period was mainly coagulase positive and sensitive to oxacillin, and hence was MSSA.

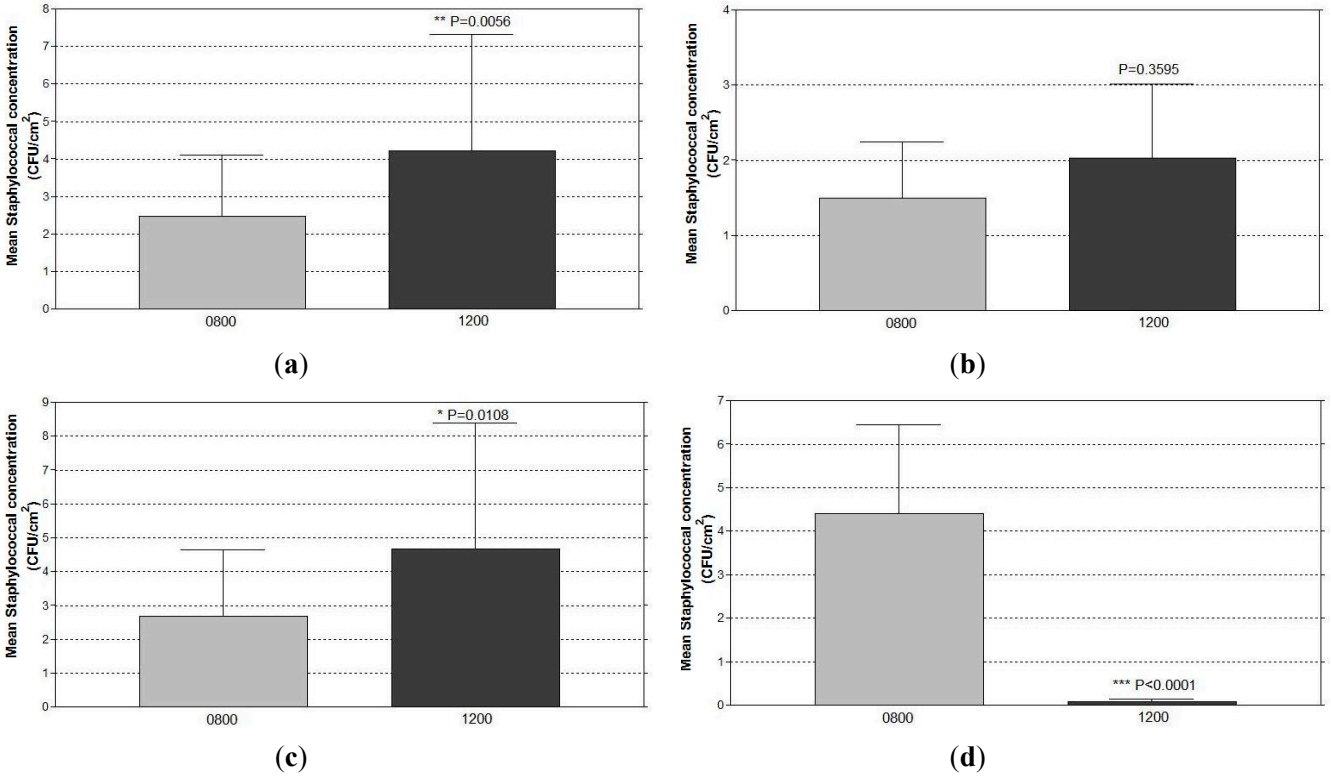
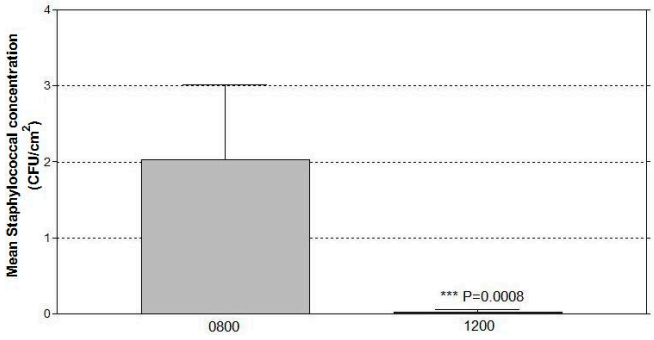



**Figure 2.** *Cont.*

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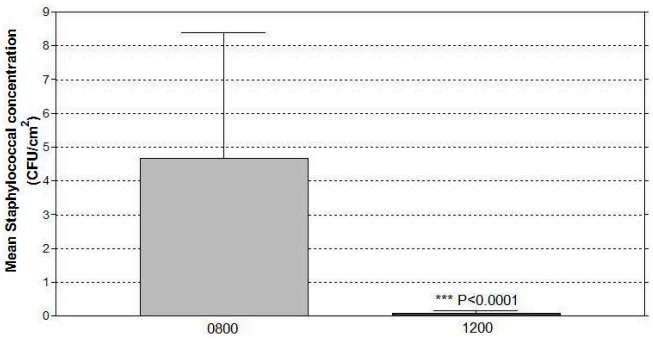


(**f**)

**Figure 2.** The increase in staphylococcal concentrations recovered from the bedside surfaces of (**a**) all control (received hypochlorite cleaning only) bed-units irrespective of the patients' MRSA status (n = 36). (**b**) MRSA-positive patients (n = 6). (**c**) MRSA-negative patients (n = 30). The reduction in staphylococcal concentrations recovered from the bedside surfaces 4 hours after the application of JUC spray of (**d**) all bed-units irrespective of the patients' MRSA status (n = 18). (**e**) MRSA-positive patients (n = 3). (**f**) MRSA-negative patients (n = 15).

With the focus on oxacillin resistance, sites of contamination were shown to be different among the bed-units of MRSA-positive and negative patients. The surface of the left-side handrail was found to be the most heavily contaminated site, containing 84% of all MRSA isolates recovered from the MRSA-negative bed-units. Indeed, the left-side handrail was found to have an MRSA concentration 11-fold significantly ($p < 0.05$) higher than that of the right-side handrail. However, with regard to the MRSA-positive bed-units, 54% of the MRSA isolates were recovered from the overbed rolling table surface as the dominant site of contamination. The mean MRSA concentration on the surface of the right-side handrail was found to be slightly higher (albeit statistically insignificant) than that on the surface of the left-side handrail, while none was recovered from the bedside table specimens.

As shown in Table 4, over the four-hour interval, the number of bed-units contaminated by staphylococci was dramatically reduced from 78% to 11% following the application of JUC polymer. Only two out of the 18 JUC-applied bed-units (11%) still tested positive for culture and all isolates were identified as CNS, while none of these bed-units had isolated positive cultures of MSSA and MRSA. In the experimental group, the mean staphylococcal concentration of the bedside surfaces had dropped significantly ($p < 0.0001$) from 4.4 ± 8.7 CFU/cm$^2$ at 08:00 hours to 0.7 ± 0.26 cfu/cm$^2$ at 12:00 hours due to the use of the QAC polymer (Figure 2d). The same trend of reduction was observed irrespective of the MRSA status of the patients (Figure 2e,f).

## 4. Discussion

The novelty of this study lies in the significant reduction in staphylococcal contamination on the bedside surfaces of the medical ward due to the use of the JUC polymer. To the best of our knowledge, this is also the first study to report on the situation of staphylococcal contamination in the general ward environment of hospitals in Hong Kong. It is also the first study to compare staphylococcal contamination between bed-units occupied by MRSA-positive and MRSA-negative patients.

**Table 4.** Number of bed-units that showed a positive growth in staphylococcal bacteria according to the staphylococcal types recovered from the specimens collected four hours after the application of JUC spray.

| Bed-Units with Positive Culture and Types of Staphylococci | No. of Bed-Units (%) | |
|---|---|---|
| | Control (Hypochlorite Cleaning) | Experimental (JUC + Hypochlorite Cleaning) |
| Bed-units of MRSA carrier (n = 3) | | |
| Positive culture | 3 (100%) | 0 (0%) |
| CNS | 1 (33%) | 0 (0%) |
| MSSA | 2 (67%) | 0 (0%) |
| MRSA | 1 (33%) | 0 (0%) |
| Bed-units of non-MRSA carrier (n = 15) | | |
| Positive culture | 11 (73%) | 2 (13%) |
| CNS | 2 (13%) | 2 (13%) |
| MSSA | 8 (53%) | 0 (0%) |
| MRSA | 3 (20%) | 0 (0%) |
| All bed-units (n = 18) | | |
| Positive culture | 14 (78%) | 2 (11%) |
| CNS | 3 (17%) | 2 (11%) |
| MSSA | 10 (56%) | 0 (0%) |
| MRSA | 4 (22%) | 0 (0%) |

Note: The experimental arm consisted of the application of JUC spray in addition to daily hypochlorite wiping, while the control arm consisted of daily hypochlorite wiping alone. A total of 18 bed-units were studied for each arm.

### *4.1. Staphylococcal Contamination on the Bedside Surfaces of Bed-Units in the Studied Ward*

Our results indicated that a large number (about 80%) of bedside surfaces was contaminated and that oxacillin resistance was prevalent, occurring not only around the beds of MRSA-positive patients but also of MRSA-negative patients, at one hour (08:00 hours) and five hours (12:00 hours) after a hypochlorite wiping (performed at 07:00 hours). The results reflected the natural contamination within the ward where hypochlorite cleaning was regarded as a routine practice, and authors are aware that it may have affected the staphylococcal identity and proportionate isolation. Surface MRSA contamination was commonly reported in bed-units or rooms occupied by MRSA infected or colonized patients [9,13,14]. With the current study design, with its particular emphasis on staphylococcal contamination in the natural clinical environment and on avoiding disturbances to routine activity and staff duties, there could have been several important sources for the MRSA contamination. First, the MRSA-positive patient could have been a stable source within the cubicle. Creamer and colleagues [21] revealed that patients could shed MRSA, more frequently early in the morning, which could spread in the surrounding air to other hospital surfaces. However, a sophisticated genotyping analysis is needed to confirm that the MRSA isolates were shed from the MRSA-positive patients and dispersed within the ward. The activities of the nursing station might also be a potential source of heterogeneous contamination. However, such activities would also be considered part of the natural ward environment. Although, in normal circumstances visitors were generally not allowed to enter the ward

during the four-hour interval, during the operation of the ward visitors could not be completely banned. Therefore, visitors and staff could also be staphylococcal carriers, and staphylococcal bacteria can survive in the environment for months [14]. Hospital staff are known to be the major vehicle of bacterial transmission during routine patient care [22,23]. Ward activities, such as ward rounds, clinical activities (resuscitation, sampling, *etc.*) and bed-making could all contribute to the environmental dispersal of staphylococci. This is consistent with the results of this study, in which the overall surface staphylococcal burden from the sampled sites increased by 80% from 08:00 to 12:00 hours, when routine ward and clinical activities were maintained. Regarding the CNS strains that were recovered from all surfaces, the most clinically important CNS causing nosocomial infections is known to be *Staphylococcal epidermidis*, which is frequently sourced from the skin normal flora of patients and healthcare workers [24]. Nonetheless, the attitudes and beliefs of the cleaning staff are considered as an additional factor that some staff may carried out the cleaning more effectively for the bed-unit of the known MRSA-positive patients. This factor cannot be ignored in the current study.

Hospital ward environments, especially high-touch surfaces, are rich reservoirs for the transmission of many microorganisms. The current results were consistent with those of previous studies reporting that handrails are the most frequently contaminated surfaces within a ward environment [14]. Handrails and overbed rolling tables were the dominant sites of contamination in the bed-units occupied by the non-MRSA and MRSA patients. The advantages of wiping high-touch surfaces daily with sporicidal agents such as bleaching water have been experimentally demonstrated [25]. Although hypochlorite disinfectants are well known for being able to eliminate a wide spectrum of bacteria, including MRSA, the current results show the drawbacks of relying on hypochlorite wipes to decontaminate hospital environments. A local study reported on the failure of disinfection efforts using hypochlorite wiping by demonstrating the presence of MRSA in bedside rails after hypochlorite wiping. A possible cause was the failure to thoroughly rinse the wipe, as suggested by the presence of bacteria in the wipe before wiping was carried out [26]. However, this study did not set out to assess the effectiveness of hypochlorite wiping. As no sampling was carried out prior to cleaning and the effectiveness of hypochlorite wiping should not be judged in current study. However, results herein suggested that one-off wiping with hypochlorite will not prevent recontamination of the surface over time. Furthermore, it is a fact that hypochlorite agents are easily inactivated by the presence of biofilm formed by bacteria and contamination by organic compounds in the hospital environment [8], and that they only offer immediate but not long-lasting antimicrobial activity, which is a non-modifiable property. Hospital wards may consider increasing the frequency with which they carry out hypochlorite wiping during the day.

### *4.2. Significant Reduction in Surface Staphylococcal Burdens from Using the Antimicrobial Coating*

The results clearly indicate that the application of JUC polymer on bedside surfaces effectively reduces both the incidence of staphylococcal contamination and bacterial concentration. The liquid preparation of JUC solidifies immediately when upon contact with the surface of skin or any fabric to form a two-sided film. The bonded film adheres firmly to the surface, and the positively charged film attracts the negatively charged cell walls and membranes of microorganisms to exert electrostatically destructive killing effects [1]. So far, *in vitro* cytotoxicity ranging from 99 to 100 percent has been

tested on pathogenic microorganisms including *Staphylococcus aureus*, *Treponena pallidum*, *Pseudomonas aeruginosa*, *Gonococcus*, *Colibacillus*, *Candida albicans*, and SARS coronavirus [27]. The results of this study suggest that the JUC polymer has a long-lasting antimicrobial activity of at least four hours after application. The manufacturer has claimed that its antimicrobial properties last for eight hours, which requires further confirmation. Regarding the toxicity, different tests were performed and JUC polymer has been proven to be safe for use directly on wounds and on the critical surfaces of medical devices [1,27]. Toxicity tests have been performed on mice and rabbits, and the lethal dose 50 ($LD_{50}$) was determined as >10,000 mg/kg, which is essentially non-toxic. In particular it has no irritation to skin and eyes [27]. These findings support the view that the JUC spray should be safe to use for environmental decontamination in hospitals.

Several researchers [28,29] have recommended the use of quaternary ammonium compound (QAC)-based antimicrobial coating on high-touch surfaces. However, a recent study has reported the occurrence of an antiseptic-resistant gene among Hong Kong nurses, which reduces the biocide susceptibility of QAC in staphylococcal organisms [30]. This aspect must be carefully addressed before the JUC spray and other QAC-based surfactants can be further implemented as an effective environmental surface decontamination aid. Despite this, as mentioned above, the antimicrobial activity of JUC spray is exerted by the physical electrostatic force generated between the positively charged coating surface and the negatively charged cell surface, which does not involve any biological or chemical mechanism that may develop the resistance. Nonetheless, the issue of resistance cannot be ignored, and the antimicrobial activity of JUC coating should also be tested against other important hospital-associated organisms such as *Pseudomonas aeruginosa* and multidrug resistant (MDR) gram-negative organisms including *Stenotrophomonas maltophilia* and vancomycin-resistant enterococci in the hospital environment. Such surface treatment should be abandoned if the MDR gram-negatives appear and cause problem for patients. To the best of our knowledge, this is the first study that has been conducted to evaluate the effectiveness of JUC spray as an enhanced surfactant disinfectant in addition to hypochlorite wiping in the general ward environment. The results were obvious on Staphylococci including the coagulase-negative and oxacillin-resistant strains, which warrants a large-scale investigation involving more hospital environments.

## 5. Conclusions

The application of JUC OrganoSiQAC-based surfactant as a antimicrobial coating was found to be effective in reducing the incidence and bacterial concentrations of bedside staphylococcal contamination. It exerted long-lasting antimicrobial activity for at least four hours after application. The finding supports the application of JUC spray as a potential environmental decontamination strategy to prevent the transmission of clinically important pathogens in medical wards.

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## Author Contributions

Terence W. K. Chung designed the study, collected the data, and wrote the first draft of the manuscript. Alice Y. Loke contributed in the finalization of the data analysis and presentation of the results. John W. M. Yuen provided guidance throughout Mr. Chung's study and wrote the final manuscript based on the first draft.

## Conflicts of Interest

The authors declare that they have no conflict of interest.

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*文章*

# 耐甲氧西林金黄色葡萄球菌（MRSA）对医院病房床边各表面的污染以及一种能提高杀菌有效性的抗微生物高分子表面活性剂

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摘要：这项研究旨在评估耐甲氧西林金黄色葡萄球菌（MRSA）阳性患者居住的常规操作的内科病房中，一种季铵盐（QAC）表面活性剂在减少表面葡萄球菌污染方面的有效性。被检测的季铵盐是一种抗菌膜，喷洒在表面能保持8小时的活性。我们设计了现场实验，实验组为季铵盐加每日次氯酸盐清洗，对照组单独采用次氯酸盐清洗。采样方法为在湿润表面用拭子采样。结果发现，08：00点时，83%的MRSA阳性患者和77%的MRSA阴性患者的床边各表面分别被葡萄球菌污染，而12：00点时，普通病房和临床上葡萄球菌的浓度在4小时之内增加了80%。不考虑患者MRSA的情况，在研究的内科病房内，尽管每天都用次氯酸盐进行擦洗，床周围高接触表面受到包括MRSA在内的葡萄球菌的严重污染（在培养结果为阳性的地方从1到276 cfu/cm$^2$不等）。然而，在使用季铵盐高分子之后，污染

率从78%显著降低至11%。在实验组，使用季铵盐高分子之后，床边各表面平均葡萄球菌浓度从08：00点时的4.4 ± 8.7 cfu/cm$^2$显著降低至12：00点时的0.07 ± 0.26 cfu/cm$^2$。实验结果支持了以下观点：联合次氯酸盐擦洗，被检测的季铵盐表面活性剂是一种潜在的消除环境污染的策略，可用于预防内科病房中临床主要病原体的传播。

**关键词:** MRSA;葡萄球菌感染；表面污染；环境污染；季铵盐；JUC；抗微生物表面活性剂

## 1. 引言

医院里各个表面的微生物都会传播到医护人员、患者和访客的手上，造成交叉感染和传染病。尽管大多数医院都执行了常规清洁和预防措施，我们仍需要有效的去除环境污染方法。近几十年以来，针对污染表面，很多高分子表面活性剂产品已被证明具有优异的抗菌性能，但没有一个在医院里的各表面上做过检测 [1，2]。JUC 喷雾剂是一种主要成分为阳离子有机硅季铵盐 （OrganoSiQAC）的纳米技术，目前是FDA 批准的用于伤口护理的隐形水凝胶抗微生物敷料。根据当地的案例报告，经证明，JUC喷雾剂也能够有效地处理 MRSA相关的皮肤脓肿 [3]。两个最近的试验 [4，5] 证明，在关键医用表面上使用 JUC 高分子能减少细菌量。这些医用表面还包括导尿管，导尿管相关感染发生率大幅减少 [5]。JUC制造商声称，这种抗微生物膜在生命体和非生命的表面上能保持长达八个小时 [1]。因此，我们的研究团队特别有兴趣调查 JUC 高分子针对医院各表面长效表面活性剂的作用，和有效辅助去污的潜在应用。

在医院里，患者密切接触的表面更有可能被污染。医院不同区域中MRSA 对环境的污染已经有大量报道，包括重症监护病房 [6-9] 、烧伤部门 [10]、隔离病房[11，12]和普通病房 [13]。在医院急性病房，在MRSA阳性患者的病房里，可以从 1%-27%的表面检测出MRSA [14]。然而，医院病房表面不同，MRSA 污染发生率也存在差异，因为污染受到各种因素的影响，如患者的情况、病房设置、是否拥挤、甚至采样方法 [15，16]。经证明，高接触表面是医院环境中MRSA的主要聚集地。在医院的所有表面中，MRSA阳性患者居住病房的床边栏杆被认为是最常被MRSA污染的地方[14]。其他经常被污染的表面包括床边曲柄、床上

桌、床上用品、床头柜、床边托盘、压力带、静脉泵、窗帘、 门把手、键盘和地板[6,9,11,13]。

医源性MRSA（HA-MRSA）感染的发生率在不同地方各有不同。香港是亚洲高发区之一[17]。根据亚洲耐药病原菌监测网（ANSORP）的研究，从香港医院住院病人中分离出的所有葡萄球菌菌株里，57%被证明是耐甲氧西林的 [18]。在香港，医疗服务是由香港政府支持的医院管理局提供的。公立医院的普通病房通常安排每个病房六张病床。不像在许多其他国家，入院时已知的MRSA定殖或感染病例将不会被隔绝或分配到单人病房。相反，这类病人会使用病房尽头的病床。由于床边的各表面环境在粘膜接触方面不被认为是关键表面，按照美国疾病控制和预防中心（CDC）的环境感染控制策略，用次氯酸盐一天清洁一次这类表面[15]。在处理MRSA患者时，采取保护性屏障的预防措施，例如使用手套和口罩。鉴于香港的MRSA高感染率和应对当地内科病房MRSA阳性患者的办法，本研究的主要目的是评估在日常内科病房里，除了每天用次氯酸盐擦拭清洁和保护性屏障的预防措施，加用JUC 高分子对床边各表面的去污有效性。据我们所知，在香港医院的普通病房迄今一直没有针对表面 MRSA 相关污染的流行病学研究。在使用JUC 高分子之前，评估内科病房内高接触床边各表面受到MRSA和其他葡萄球菌污染的程度，尤其是MRSA阳性患者居住的病房。

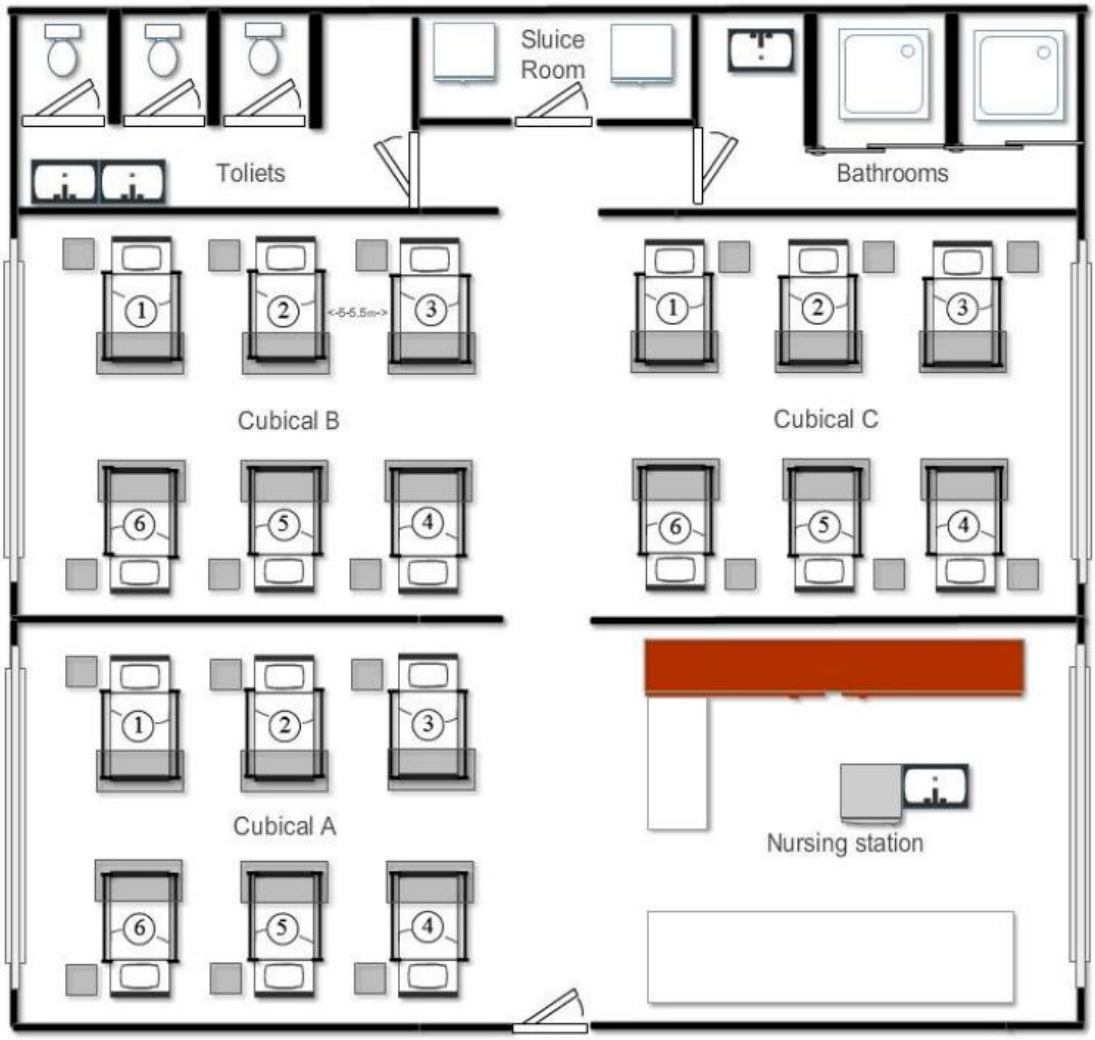
## 2. 实验部分

现场实验研究旨在评估内科病房床边各表面上葡萄球菌污染的程度，以及 JUC 高分子减少这种污染的效果。在病房里继续日常的操作，以反映最自然的环境，从而进行评估。

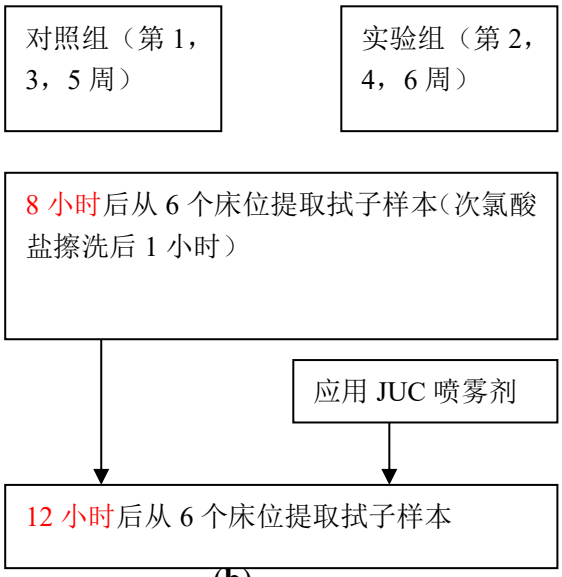
### *2.1.研究医院病房的设置*

研究医院是香港一家大型的教学医院，有 1500 张病床。研究病房为男性内科病房，由三个病房组成，每个病房有 6 张病床，在研究期间病床每天都全满。病房和病房设置见图 1a。A 病房被指定为这项研究所用的病房。每个病房为 53 乘 60 平方米，每个床位为 21 乘 9 平方米，床间距离为 5 至 5.5 米。在病房里保持 24.5 至 25.5 摄氏度的室内温度和 50%到 55%的湿度。病房里有适当的通风系统，24 小时空调，所有窗户都是关闭的。只有当病房住有已知 MRSA 感染或定殖时间

不少于 24 小时的患者时，才进行抽样，以评估污染和实验来评价 JUC 高分子。按照惯例，把病房里一号床分配给 MRSA 阳性患者（图 1a）。




(a)




**(b)**

**图 1(a)** 三个病房的病房布局，每个病房由六个床位组成。在 A 病房进行实验和环境取样，因为它与其他两个病房隔离开来，相对远离闸室、浴室和厕所。在采样的每一天，MRSA 患者都使用病房尽头的床位 1。**(b)** 实验组和对照组的实验计划，说明整个实验需要 6 周时间，因为每次采样后有一周冲洗时间，从而避免 JUC 喷雾剂的携带污染因素 （如果有的话）。

*2.2.评估床边表面上的金黄色葡萄球菌污染*

选择四个高接触床边表面部位（包括床边桌子、左侧扶手、右侧扶手和 床上滚动桌）用于拭子采样。采用定量方法 确定所有选定的部位为高接触表面 [19]。主要研究开始前，测试采样方法。试验结果还表明：所有所选部位均存在很大程度污染（发现 83%的培养表面标本呈金黄色葡萄球菌阳性， 每个采样部位平均范围 1-27 CFU/cm$^2$）。发现所用的尼龙植绒或大泡海绵拭子是一种从外界环境表面获得细菌的可靠有效方法[20]。在此研究中，采用“湿润表面棉签擦拭”的方法，Puritan ® 无菌大泡采样棒环境拭子涂药器喷头用于拭子采样。在每个采样表面（标准 10 cm$^2$）的三个随机点进行棉签擦拭。因此， 从每个床位共收集到 12 个拭子标本 （每个部位 3 个样本×4 个点）。棉签擦拭的标本被接种至含有 1mL 无菌水的小瓶中，再转移到 Petrifilm TM 金黄色葡萄球菌表达计数平板 上（3M 公司，圣保罗，明尼苏达州，美国）。

将 MRSA 阳性和 MRSA 阴性患者占用床位的污染程度进行比较。在 08：00 点和 12：00 点两个时间点，从病房内所有床位收集拭子标本，然后在 6 周期间 6 个独立的日子重复上述操作。选择上午 8 点的时间点是因为，在 07：00 点次氯酸盐擦拭后时间不是很长，多数患者在病房内夜班的时候活动量最少，然后醒来。责任医护人员根据医院指南制备新鲜的次氯酸溶液，同时在责任护士的监督下擦拭整个病房以确保所有表面干净。因此，每个 08：00 点标本被用作基线进行 MRSA 阳性和 MRSA 阴性表面之间的比较。此外，每个 12：00 点标本被用来评估不受干扰情况下进行常规临床活动的四小时间隔期内污染的自然变化。在午餐前四小时期间，通常不允许探视人员进入病房，以最大程度地减少除患者和医务工作者以外的干扰。然而，不能阻止病房外的医护人员进入病房。经过 6 周的研究，总共从指定的部位表面收集 864 个拭子标本 （每个部位 3 个样本×4 个点×6 张床 × 2 个时间点× 6 天）。

*2.3 实验设计用于评估 JUC 高分子的去污作用*

在这个实验中，相同病房相同指定床位进行 JUC 高分子对金黄色葡萄球菌的去污性能评估。实验组测试 JUC 喷剂加常规每日清洁（即在每个 07：00 点用漂白水消毒），同时对照组仅测试常规每日清洁。所有研究组，收集每个 08：00 点的拭子标本作为基线，收集每个 12：00 点的拭子标本用于评估金黄色葡萄球菌负荷的变化。JUC 喷雾敷料由南京神奇科技开发有限公司（中国南京市）捐赠。在实验期间，收集基线标本（见图 1b）后立即 JUC 喷雾剂应用于部位表面。制造商推荐以下喷雾方法：在距离 10 cm 处均匀地喷洒目标表面 5 秒以覆盖点的区域。喷洒时不应产生危险的气雾，因为 JUC 喷雾剂 98%是由水构成。JUC 喷雾剂应用于病房内所有床位上，这个过程在 3 周内重复 3 次，总共 72 个喷洒点，即每张床 4 个部位×每个病房 6 张床×实验 3 天。因为从每个表面部位收集了三个随机拭子，每个实验组或对照组共有 216 个标本。棉签擦拭的标本接种至含有 1 mL 无菌水的小瓶中，并将其转移至 Petrifilm TM 金黄色葡萄球菌表达计数平板（3M 公司）上。为了避免 JUC 喷雾剂携带污染，将每次实验天数安排为至少 7 天，除了实验次日。对照组在单独的三天进行试验。在每个实验和对照日，一例 MRSA 阳性患者在病房的尽头使用床位，即图 1 中的床位 A1。病房工作人员

和医务人员在实验中采用盲法以确保常规病房活动不受影响和干扰。使用床位的患者的人口特征，包括其 MRSA 状况，总结于表 1 中。

表 1：在每个实验和对照日，使用床位的患者的人口特征

| 参数 | | 试验组 | 对照组 | 患者 t 检验 |
|---|---|---|---|---|
| 平均年龄±标准差（范围） | | 81.13 ± 7.17 (67-91) | 73.44 ± 16.26 (38-92) | 0.1041 |
| 患者总数 | | 18 | 18 | 不适用 |
| 从敬老院收入的（患者数量） | 是 | 11 (61%) | 10 (63%) | 不适用 |
| | 否 | 7 (39%) | 6 (37%) | 不适用 |
| MRSA 阳性培养#（患者数量） | 伤口拭子 | 1 | 1 | 不适用 |
| | 鼻拭子 | 0 | 1 | |
| | 痰 | 2 | 1 | 不适用 |
| | 无 | 15 | 15 | 不适用 |
| 平均住院天数±标准差（范围） | | 7.81 ± 4.35 (2-16) | 6.75 ± 2.95 (1-14) | 0.4395 |

备注：总共 18 例患者（含单独的三天内 3 例 MRSA 阳性患者）被纳入研究的各个组（试验组：JUC 喷雾剂+标准清洁，对比，对照组：标准清洁）。在单独的三天内（每次间隔 7 天）对患者进行研究。每天，所有 6 张病床都被使用，使用者包括病房小隔间内 1 例 MRSA 携带者和 5 例非 MRSA 携带者。

*2.4 细菌培养和鉴定*

收集样品一小时内, 所有接种的金黄色葡萄球菌表达计数平板被送至研究医院的微生物学实验室培养和进一步分析。所有平板在在 37℃下培养 24-48 小时。紫红色菌落被计数为金黄色葡萄球菌生长阳性，并以每平方厘米集落形成单位（cfu/$cm^2$）表示。因为从 10 $cm^2$ 的表面积收集每个拭子样品，CFU/Petrifilm 实际上代表了每份 10 $cm^2$ 表面积拭子标本上金黄色葡萄球菌菌落的生长情况。采样技术的最低（可计数）检出限被定义为 1 cfu/$cm^2$。使用凝固酶试验，跟踪平

板上的阳性细菌生长，鉴别凝固酶阳性和凝固酶阴性葡萄球菌（CNS）。用 Mueller-Hinton 琼脂辅以 4% w/v 氯化钠继代培养凝固酶阳性葡萄球菌（从金黄色葡萄球菌表达计数平板随机选取紫红色菌落），按照美国疾病控制和预防中心（http://www.cdc.gov/HAI/settings/lab/lab_mrsa.html）的建议，采用纸片扩散法评估其对苯唑西林的敏感性。MRSA 菌株对所有β-内酰胺类抗生素耐药，但通常耐苯唑西林。因此，在本研究中，耐甲氧西林葡萄球菌金黄色葡萄球菌（MRSA）菌株被鉴定，并与甲氧西林敏感葡萄球菌金黄色葡萄球菌（MSSA）菌株区分。根据 JUC 喷雾敷料的技术报告和使用说明，季铵盐（QAC）溶液喷洒在任何表面上与空气接触后立即固化，在带正电荷的涂层表面和带负电荷的微生物细胞壁或细胞膜之间产生的物理静电力作用下发挥杀菌性能。表面附着的涂层将具有防水性，并能保持 8 小时作用。擦拭棉签时，不应从采样表面清除该涂层。此外，如果该涂层在溶液中释放，物理杀菌活性将丢失，因此在从有 JUC 涂层的床头表面标本收集程序中不需要使用中和剂，残余季铵盐（QAC）不会影响微生物的结果。

*2.5.道德方面的考虑*

实验程序由香港理工大学和研究医院的伦理委员会 审查和批准。本研究仅涉及病房隔间内床位周围环境的表面，但不涉及患者和病房工作人员。JUC 喷雾剂是 FDA 批准的伤口护理产品，不会对人体产生伤害。入院时，根据患者的病史和医院感染控制小组提供的信息证实患者的 MRSA 状况，此信息未在任何地方向任何人披露。

**3. 结果**

08:00点时，从所研究病房内83% (5/6) 的MRSA阳性患者床头表面和77% (23/30) 的MRSA阴性患者床头表面获取阳性葡萄球菌（表2）。无论MRSA携带者状态如何，从所有床位获取凝固酶阳性葡萄球菌 (CPS) 和凝固酶阴性葡萄球菌 (CNS)。08:00点（次氯酸盐清洗后一小时）时，从各床位的床头表面采集三个随机拭子标本，所以每个床头表面共检测108个拭子。采样的床头表面大部分细菌生长呈阴性（表3总结如下：CNS阳性率为4.6%–11.1% ；MSSA阳性率为2.8%–9.3% ；MRSA 阳性率为7.4%–17.6%）。细菌生长呈阳性的那些表面，CNS、MSSA和MRSA恢复的菌落数范围分别为1–16、1–193和 1–276 cfu/cm$^2$。不考虑

拭子涂抹部位的情况下，所有采样床位中分别有44%、28%和56%获取得到CNS、MSSA和MRSA。MRSA阳性患者和MRSA阴性患者床位的平均CNS浓度分别为1.9 cfu/$cm^2$ 和 1.6 cfu/$cm^2$。在MRSA阳性患者床位，发现CPS 和 CNS比率为2:4，所有CPS 分离菌都对苯唑西林有耐药性，整体浓度为3.9 cfu/cm2。另一方面，从MRSA阴性床位来看，CPS分离菌是MSSA和MRSA的混合物。一半的 CPS分离菌显示对苯唑西林有耐药性，平均浓度为7.9 cfu/cm2，这明显比MRSA阳性床位获得的数量要高 ($p < 0.05$)。总的来说，CNS， MSSA ，和MRSA中的葡萄球菌分布比率为2:6:8。从受测病房格局来看（图1a），MRSA阳性床位旁边的床位细菌生长更严重（即床位2 和6）。床位 4 的细菌生长也相对较多。但与气体床位相比都没有统计学差异（数据未表明有统计学差异）。在只使用次氯酸盐清洗的所有对照床位中，从8点到12点，平均葡萄球菌污染显著增加($p < 0.01$) 了80%（图2a）。

**表2.** 08:00点时在MRSA阳性患者和MRSA阴性患者床位床头表面获得的葡萄球菌浓度及种类比较

| **葡萄球菌种类及采样部位** | **平均 cfu/cm2 ±平均标准误差 (SEM)** | | |
|---|---|---|---|
| | **MRSA阳性床位(n = 6)** | **MRSA阴性床位 (n = 30)** | **P值** |
| CNS | | | 0.1953 |
| 床头柜 | 未检出 | 0.04 ± 0.03 | |
| 左侧扶手 | 0.39 ± 0.33 | 0.12 ± 0.09 | |
| 右侧扶手 | l 1.94 ± 1.13 | 1.20 ± 1.13 | |
| 床上滚动桌 | 未检出 | 0.29 ± 0.25 | |
| MSSA | | | 未确定 |
| 床头柜 | 未检出 | 0.48 ± 0.39 | |
| 左侧扶手 | 未检出 | 1.94 ± 1.83 | |
| 右侧扶手 | 未检出 | 3.08 ± 2.17 | |
| 床上滚动桌 | 未检出 | 0.02 ± 0.01 | |
| MRSA | | | 0.0392 |

| | | | |
|---|---|---|---|
| 床头柜 | 未检出 | 0.68 ± 0.63 | |
| 左侧扶手 | 0.72 ± 0.72 | 6.37 ± 4.03 | |
| 右侧扶手 | 1.11 ± 0.98 | 0.57 ± 0.30 | |
| 床上滚动桌 | 2.11 ± 1.41 | 0.28 ± 0.14 | |

注：在6个不同的日子，于08:00点对共36个床位进行采样。所研究病房在这些天都住满人，每个采样的日子只有1位MRSA阳性患者。MRSA阳性和MRSA阴性患者之间MSSA 数量的统计学差异不能确定，因为一个比较组的计数为0。N.D. = 未检出，说明petrifilm 板上不存在菌落。采样技术的检测限值为1 cfu/Petrifilm（相当于cfu/$cm^2$）。由于该表中有大量0值（未检出），所以该表中的平均 cfu/$cm^2$ 值小于检测限值（有关各采样部位的阳性培养菌比例，请参见表3）。

**表3.** 08:00点时（次氯酸盐清洗后一小时），从表面擦拭部位获取的三种葡萄球菌的数量，百分比以及菌落数范围

| **葡萄球菌的类型和参数** | **表面擦拭部位（各部位 n = 108）** | | | |
|---|---|---|---|---|
| | **床头柜** | **左侧扶手** | **右侧扶手** | **床上滚动桌** |
| CNS | | | | |
| 阳性培养菌数量(%) | 5 (4.6) | 6 (5.6) | 12 (11.1) | 7 (6.5) |
| 阳性培养菌中的cfu/cm2范围 | 1–2 | 1–9 | 1-16 | 1-14 |
| MSSA | | | | |
| 阳性培养菌数量(%) | 7 (6.5) | 10 (9.3) | 4 (3.7) | 3 (2.8) |
| 阳性培养菌中的cfu/cm2范围 | 1-36 | 1-164 | 1-193 | 1-2 |
| MRSA | | | | |
| 阳性培养菌数量(%) | 8 | 19 (17.6) | 19 | 12 (11.1) |

| | | | | |
|---|---|---|---|---|
| 阳性培养菌中的cfu/cm2范围 | (7.4)<br>1-23 | 1–276 | (17.6)<br>1–75 | 1-26 |

MRSA阳性患者和阴性患者床位中观察到的趋势一样；但MRSA阴性床位的葡萄球菌污染增加值（从2.8 升至4.8 cfu/cm2），约为MRSA阳性床位中增加值（从1.5升至2.0 cfu/cm2）的4倍（图2b,c）。4小时后增加的葡萄球菌菌株主要是凝固酶阳性，并且对苯唑西林敏感，因此是MSSA。

图2：

葡萄球菌平均浓度

图2. 无论患者MRSA状态如何(n = 36)，(a)所有对照（只使用次氯酸盐清洗）床位的床头表面获取的葡萄球菌浓度上升情况；(b) MRSA阳性患者(n = 6)，床头表面获取的葡萄球菌浓度上升情况；(c) MRSA阴性患者(n = 30) 床头表面获取的葡萄球菌浓度上升情况。(**d**) 无论患者MRSA状态如何(n = 18)，使用JUC喷剂4小时后，所有床位床头表面获取的葡萄球菌浓度下降情况； (**e**) MRSA阳性患者(n = 3) ，床头表面获取的葡萄球菌浓度下降情况；(**f**) MRSA阴性患者(n = 15) 床头表面获取的葡萄球菌浓度下降情况。

以苯唑西林耐药性为重点，MRSA阳性患者和阴性患者床位的污染部位不同。研究发现，左侧扶手的表面是污染最严重的部位，占MRSA阴性床位中所有MRSA分离菌的84%。事实上，左侧扶手MRSA浓度比右侧扶手浓度显著高出11倍($p < 0.05$)。但对于MRSA阳性床位，54%的MRSA分离菌来自床上滚动桌，是污染的主要部位。右侧扶手表面的平均MRSA浓度略高于（虽然统计上不显著）左侧扶手表面，但床头柜上没有检测出MRSA。

如表4所示，应用JUC高分子，间隔4小时后，受到葡萄球菌污染的床位数量显着地从78%下降到11%。18个应用 JUC的床位只有2例 (11%)仍检测到阳性培养菌，并且所有分离菌都鉴定为CNS，这些床位均没有分离出MSSA 和 MRSA阳性培

养菌。在实验组，由于使用了有机硅季铵盐高分子，床头表面的平均葡萄球菌浓度从8点时的4.4 ± 8.7 CFU/cm2显著下降到12点时的0.7 ± 0.26 cfu/cm2（图2d）。不管患者MRSA状态如何，都呈同样的下降趋势（图2e,f）。

## 4. 讨论

本研究的新颖之处在于，使用JUC高分子后，内科病房的床头表面葡萄球菌污染显著减少。据我们所知，这也是报告香港医院普通病房环境下金黄色葡萄球菌污染的第一项研究。这也是比较MRSA阳性患者和MRSA阴性患者之间葡萄球菌污染情况的首次研究。

**表4.** 应用JUC4小时后，各葡萄球菌类型中葡萄球菌呈阳性的床位数

| **阳性培养菌的床位数及葡萄球菌类型** | **床位数 (%)** | |
|---|---|---|
| | **对照组（次氯酸盐清洗）** | **实验组（JUC+次氯酸盐清洗）** |
| MRSA携带者床位(n = 3) | | |
| 阳性培养菌 | 3 (100%) | 0 (0%) |
| CNS | 1 (33%) | 0 (0%) |
| MSSA | 2 (67%) | 0 (0%) |
| MRSA | 1 (33%) | 0 (0%) |
| 非MRSA携带者床位(n = 15) | | |
| 阳性培养菌 | 11 (73%) | 2 (13%) |
| CNS | 2 (13%) | 2 (13%) |
| MSSA | 8 (53%) | 0 (0%) |
| MRSA | 3 (20%) | 0 (0%) |
| 所有床位(n = 18) | | |
| 阳性培养菌 | 14 (78%) | 2 (11%) |
| CNS | 3 (17%) | 2 (11%) |

| | | |
|---|---|---|
| MSSA | 10 (56%) | 0 (0%) |
| MRSA | 4 (22%) | 0 (0%) |

注：实验组在每日涂抹次氯酸盐之外加喷JUC，而对照组每日只使用次氯酸盐。每组共有18个床位。

*4.1 实验病房床头表面的金黄色葡萄球菌污染*

研究结果显示，床头表面大部分（约 80%）受到污染了，苯唑西林耐药性十分普遍，在用次氯酸盐擦拭以后（7 点钟进行）1 小时（8 点钟）和 5 小时（12 点钟），不止 MRSA 阳性患者的床头被污染，MRSA 阴性患者的床头也有污染。这些结果反映出，虽然病房里会进行常规的次氯酸盐清洗，但仍有自然污染，作者认为这可能会影响金黄色葡萄球菌的个性和相应的分离。表面 MRSA 感染通常报告存在于 MRSA 感染的病房、病床或携带患者身上[9,13,14]。在当前的实验设计中，特别强调自然临床环境下的金黄色葡萄球菌污染，并且避免干扰日常活动和工作人员的职责，我们发现了 MRSA 污染的几个非常重要的来源。首先，MRSA 阳性患者可能是小隔间内 MRSA 污染的稳定来源。Creamer 和他的同事指出，MRSA 通常会在清晨从患者身上释放出，然后随着周围的空气传播到其他医院表面。但是需要一个先进的基因分型分析来证明 MRSA 能从阳性患者身上释放出，并在病房里传播。护士站的活动也可能是异构污染的一个潜在来源，但是这种活动也可以看做是自然病房环境的一部分。虽然在正常情况下，一般不会允许访客在四小时的间隔内进入病房，但是在病房的正常运行过程中访客是不能完全被禁止的。因此，访客和工作人员也有可能是金黄色葡萄球菌携带者，而且金黄色葡萄球菌可以在环境中存活数月。医院的工作人员已知为日常护理病人中细菌传播的主要载体。病房活动，如查房，临床活动（复苏术，采样等）和铺床等都可能导致金黄色葡萄球菌在环境中的扩散。这与本研究的结果是一致的，在维持常规的病房和临床活动的情况下，从 8 点到 12 点钟，采样点的整个表面金黄色葡萄球菌负载量增加了 80%。而对于所有表面上恢复的 CNS 菌株，临床上引起院内感染的最重要的 CNS 已知为表皮葡萄球菌，通常来自于患者和医护人员皮肤表面的正常菌群[24]。虽然如此，保洁人员的态度和信念也被认为是一个额外的因素，对于某些

已知的 MRSA 阳性患者，保洁人员会更有效率地清洁他们的病床。在当前的研究中，这个因素不能忽视。

医院病房环境，特别是高度接触的表面，是众多微生物传播的发源地。之前的研究报告称扶手是病房环境中一个最常被污染的表面，我们的结果也是这样。MRSA 患者和非 MRSA 患者所用的病床中，扶手和床上桌是污染的主要部位。实验证明每天使用杀菌剂，如漂白水，来擦拭高度接触的表面是有好处的。虽然次氯酸盐消毒剂公认为能广谱抗菌，这其中也包括 MRSA，但是目前的结果显示出仅依靠次氯酸盐擦拭来净化医院环境是不足的。当地的一个研究指出次氯酸盐擦拭完毕之后，MRSA 在床栏上仍然存在，报告了使用次氯酸盐擦拭来进行消毒工作是不成功的。一个可能的原因是未能彻底的冲洗擦布，因为在擦拭进行之前擦布上就有细菌存在[26]。不过，这项研究并没有评估次氯酸盐擦拭的有效性。由于在清洁之前没有进行取样，所以本研究也不应评判次氯酸盐擦拭的有效性。但是此处的结果能说明，一次性次氯酸盐擦拭不能预防一段时间后表面的二次污染。另外，如果细菌形成了生物膜，或者在医院环境中受到有机物污染，次氯酸盐制剂就很容易失活，而且仅能提供即时而非长效的抗菌作用，这是一个不可更改的属性。医院病房可考虑增加每天进行次氯酸盐擦拭的频率。

*4.2 使用抗微生物膜以后表面金黄色葡萄球菌负载量显著减少*

结果清楚地表明， 在床头表面使用 JUC，有效地降低了金黄色葡萄球菌污染的发生率和细菌浓度。JUC 的液体制剂接触到皮肤或任何织物表面，立即固化形成一层双面膜。胶联层紧紧附于表面，而正电荷层吸附带负电荷的病原微生物，通过静电作用破坏细胞壁和细胞膜，从而实现抗菌效果[1]。目前，已检测出对病原微生物的体外细胞毒性范围为 99%-100%，包括金黄色葡萄球菌，梅毒螺旋体，铜绿假单胞菌，淋球菌，大肠杆菌，白色念珠菌和 SARS 冠状病毒[27]。研究的结果表明， JUC 在使用后至少有 4 小时的长效抗微生物活性。制造商称其抗菌性能持续八个小时，这需要进一步证实。至于毒性，进行了不同的检测，证明 JUC 是安全的，可以直接用于伤口和医疗器械的关键表面[1,27].利用老鼠和兔子进行了毒性试验，致死剂量 50（ LD 50 ）被确定为> 10,000 毫克/千克，实质上是无毒的。特别是它对皮肤和眼睛没有刺激[27] 。这些结果支持了这样的

观点：JUC 喷雾能安全用于医院环境的净化。

一些研究员[28,29]已经推荐在高度接触的表面使用季铵盐化合物(QAC)系的抗微生物膜。但是最近的一个研究报告称在香港的护士中发现了一种杀菌剂抗性基因，能减少 QAC 对金黄色葡萄球菌的抗菌敏感性[30]。必须先认真解决了这个问题，才能进一步将 JUC 喷雾和其他 QAC 系表面活性剂确定为有效的环境表面去污剂。尽管如此，如上所述，JUC 喷雾的抗菌效果是通过正电荷涂层和负电荷细胞表面的物理静电作用实现的，没有涉及任何生物学或化学机制，不会产生抗药性。然而，抗药性的问题也不容忽视，也应当用其他重要的医院相关微生物来测试 JUC 的抗微生物活性，如铜绿假单胞菌，以及多重耐药（MDR ）的革兰氏阴性微生物，包括医院环境中的嗜麦芽窄食单胞菌和耐万古霉素肠球菌。如果 MDR 革兰氏阴性菌出现并对患者产生影响，那这种表面处理的方式就应该放弃。尽我们所知，我们的研究首次评估了在普通病房环境中，除了次氯酸盐擦拭，JUC 作为一种加强的表面活性杀菌剂的有效性。结果对于葡萄球菌是显而易见的，包括凝固酶阴性和苯唑西林耐药菌株，值得在更多的医院环境中进行大规模研究。

## 5.结论

我们发现应用JUC有机硅季铵盐表面活性剂作为抗菌涂层能有效减少病床金黄色葡萄球菌污染的发生率和细菌浓度。JUC使用后能发挥至少4小时长效抗菌作用。这些结果支持JUC作为一种有潜力的环境净化策略来预防病房内临床重要致病菌的传播。

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## 作者贡献

Terence W. K. Chung设计了该实验，收集了数据并撰写了第一稿。Alice Y. Loke完成了数据分析的定稿和结果陈述。John W. M. Yuen对钟先生的研究提供

了全程指导，并在初稿的基础上完成了终稿。

## 利益冲突

作者宣称没有利益冲突。