Impact of environmental cleaning on the colonization and infection rates of multidrug-resistant Acinetobacter baumannii in patients within the intensive care unit in a tertiary hospital

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Research

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Abstract

Objective: To continuously evaluate the effect of environmental cleaning on the colonization and infection rates of multidrug-resistant *Acinetobacter baumannii* (MDR-AB) in the patients within an intensive care unit (ICU).

Methods: Environmental cleaning on the high-touch clinical surfaces (HTCS) within a comprehensive ICU was evaluated through monitoring fluorescent marks when the overall compliance with hand hygiene during 2013-2014 was monitored. Meanwhile, samples from the HTCS and inpatients were collected and sent for bacterial culture and identification. The drug susceptibility testing was further implemented to monitor the prevalence of MDR-AB. The genetic relatedness of MDR-AB collected either from the HTCS or inpatients was analyzed by pulsed field gel electrophoresis (PFGE) when an outbreak was doubted.

Results: The overall compliance with hand hygiene remained relatively stable during 2013-2014. Under this circumstance, the clearance rate of fluorescence marks on the environmental surfaces within ICUs significantly increased from 21.9% to 85.7%, and accordingly the colonization and infection rates of MDR-AB decreased from 16.5‰ to 6.6‰ and from 7.4‰ to 2.8‰, respectively, from the beginning to the end of 2013. However, during the year 2014, because of frequent change and movement of cleaning workers, the clearance rate of fluorescence marks decreased below 50%, and the overall colonization and infection rates of MDR-AB correspondingly increased from 9.1‰ to 11.1‰ and from 1.5‰ to 3.9‰, respectively. PFGE displayed a high genetic relatedness between the MDR-AB strains analyzed, indicating a dissemination of MDR-AB during the surveillance period.

Conclusion. For the easily disseminated MDR-AB within ICUs , the clearance rates of fluorescence labeling on HTCS is negatively corelated with the hospital infection rates of MDR-AB. Such an invisible fluorescence labelling is an effective and convenient method to continuously monitor cleanness of medical environment within hospitals.

Introduction

Healthcare-associated infection (HAI) is a global problem for patients, especially those inpatient with immunocompromised or critically ill diseases, causing extended hospital stays, high costs, and high mortality (1). Epidemiological studies showed that HAI is closely associated with microbial pathogens, such as methicillin-resistant *Staphylococcus aureus* (MRSA) (2, 3), vancomycin-resistant *Enterococci* (VRE) (3), and multidrug-resistant Gram-negative bacilli (4), which could be spread by the hospital environment surfaces (5). In USA and Europe, MRSA and VRE are the main pathogens associated with HAI within ICUs (3, 6, 7). In contrast, in China, the prevalence of multidrug-resistant *Acinetobacter baumannii* (MDR-AB) greatly exceeds both MRSA and VRE (8, 9), and the frequent expansion of MDR-AB poses tough challenges to control HAIs, especially those occurred in ICUs (10) (11). *A. baumannii* possesses the super survivability on kinds of healthcare equipment surfaces, from 5 days to more than 5 months (12), which surely increases the chance of transmission of MDR-AB. In addition, such strain is

difficult for prevention once it acquires resistance to the conventional detergents and alcohol disinfectants (13). The worrying condition is, the current arsenal to target MDR-AB is almost exhausted (14). Therefore, management of *A. baumannii* clusters in hospitals is the very important part of eradication of multidrug-resistant infections. The targeted infection control measures of MDR-AB including intensive cleaning and subsequent measurement of cleanliness are imperative to prevent the nosocomial acquisition and further dissemination.

The guardrail, bedside table, injection pump button, monitor button, treatment vehicle and treatment table were chosen and referred to as HTCS (high-touch clinical surfaces, HTCS), basing on hand contact frequency and easy-to-contact area of patients, which was specifically recommended in 2002 by the Centers for Disease Control and Prevention (CDC)(15). To date, it's known that the patient care items and HTCS within hospital wards are blamed for pathogen transmission (15), and the cleaning and disinfection of HTCS are pivotal for prevention of outbreak of MDR pathogens. Thus, the HTCS are always the focus of intensive cleaning in high-risk areas, especially when MDR-AB is epidemic or endemic. Considering the fluorescent marker does not allow for direct assessment of the degree of disinfection, previous studies showed that an invisible fluorescent marker is a much better strategy to improve environment cleaning, by quantitatively assessing cleaning and disinfecting practices on MRSA and VRE (16, 17). However, to our best knowledge, there is no report on this method used to evaluate impact of hospital environmental cleaning in China, especially for MDR-AB.

In this study, we utilized an invisible fluorescent marker, together with bacterial culture and identification, to evaluate the environmental cleaning on the HTCS within a comprehensive ICU of Nanjing Drum Tower Hospital, and to explore the impact of environmental cleaning on the colonization and infection rates of MDR-AB in patients from 2013 to 2014. In addition, the relationship between colonization rates and infection rates was also analyzed.

Materials And Methods

Study design

The study was conducted within Nanjing Drum Tower Hospital, a 3,325-bed general tertiary care and university-affiliated teaching hospital in Nanjing, Jiangsu province, China. Ethical approval was approved by the Ethics Committee of Nanjing Drum Tower hospital (Number: 2013-042).

The study design was shown in Figure 1. Briefly, the samples of patients hospitalized into in a 27-bed intensive care unit (ICU) in our hospital were taken to monitor MDR-AB from 2013 to 2014. The HTCS were marked by fluorescence labeling and continuously monitored for clearance level of fluorescence labeling and for contamination rate of MDR-AB, which was resistant against at least one agent in three or more of tested antimicrobial categories (18).

Investigation on the hand hygiene compliance

Observational survey of compliance with hand hygiene were conducted. Healthcare personnel was required to perform hand hygiene in accordance with the guidelines recommended by Centers for Disease Control and Prevention (CDC) (19). An alcohol-based hand rub or wash with soap and water should been used according to following the special indications for hand hygiene (20). The potential opportunities for hand hygiene were recorded according to recommended guidelines (21-23), and the actual number of episodes of handwashes and hand rubs were also noted. 20-min observations were conducted at optional time periods throughout the week. Healthcare personnel did not know the schedule of observation periods. The observers were as unobtrusive as possible, but were not hidden.

Labeling of fluorescence marks and determination of the clearance level

The fluorescence marks were drawn by a fluorescent pen (RUHOF), which uses a special nontoxic target solution. When exposed to black light, the marks emit fluorescence brightly. Noteworthily, the fluorescence marks are inconspicuous, dry rapidly on surfaces, remain environmentally stable for several weeks, resist dry abrasion, but could be easily removed by minimal abrasion with moistened cloth (17, 24). Fluorescence labeling was performed twice a day before cleaning (at 10 a.m. and 5 p.m. respectively), one mark every square centimeter. The targets were evaluated after the twice daily routine cleaning. Terminal cleaning was performed after inpatients were transferred from the ICUs. Clearance level of fluorescence labeling was calculated by comparing the number of fluorescence marks before and after cleaning. Environment cleanliness was divided into cleaning (labeling clearance rate >80%) and contamination (labeling clearance rate < 80%).

Monitoring contamination of MDR-AB on the HTCS

To monitor contamination of MDR-AB on the HTCS of the ICUs, before the daily cleaning was performed, samples were accordingly taken for bacterial culture from each site of fluorescence labeling by a cotton swab moistened with saline, according to the Technical Specification for Disinfection of Hospital Disinfection Hygiene Standard issued by the Ministry of Health of China (http://www.biaozhun8.cn/biaozhun108760). Once *A. baumannii* was detected, antimicrobial susceptibility was further implemented to detect MDR-AB.

The colonization and infection rates of MDR-AB among inpatient in the ICU

Clinical samples including sputum, urine, blood, etc., from patients within the ICU during 2013-2014 were routinely taken and sent to the clinical microbiology laboratory for bacterial culture and susceptibility testing once infections were suspected. The diagnostic criteria for colonization and infection referred to the criteria issued by the US CDC in 2008 (25). According to the international epidemiological quantitative statistical methods, the newly isolated multidrug-resistant bacteria per thousand bed days was adopted as the quantitative statistical standard, that is, the detection or infection density of multidrug-resistant bacteria in a specific time range (Number of newly isolated multidrug-resistant bacteria infected or colonized new patients in a period/number of hospital days in a period).

Bacterial identification and antimicrobial susceptibility testing

Strains isolated were identified by ATB32E or Vitek-2 technology (BioMerieux, France). The susceptibility was determined by Kirby-Bauer method. The tested antimicrobial agents were as follows: amikacin, ceftazidime, cefoperazone/sulbactam, imipenem, meropenem, piperacillin-tazobactam, cefepime, ticarcillin/clavulanate, ciprofloxacin, levofloxacin, sulfamethoxazole, minocycline and tigecycline. *Escherichia coli* American Type Culture Collection (ATCC) 25922 and *Pseudomonas aeruginosa* ATCC27853 were used as the quality controls in parallel. The results were interpreted according to guidelines of Clinical Laboratory Standard Institute (CLSI) 2015(26). However, the interpretation of tigecycline was referred to the guidelines of the current European Committee on Antimicrobial Susceptibility Testing (EUCAST) (www.eucast.org), cutoff MICs of ≤1 μg/ml and >2 μg/ml were used for tigecycline as the susceptibility and resistance breakpoints, respectively.

Pulsed field gel electrophoresis

When 14 MDR-AB isolates were detected simultaneously during Jan-Mar, 2013, the genetic relatedness among those MDR-AB strains collected from the patients and the HTCS during the same period were further analyzed through pulsed field gel electrophoresis (PFGE) according to the protocol (27). Briefly. Fresh and pure bacterial cultures were embedded in agarose plugs and digested with proteinase K (20 mg/mL), followed by *pal* restriction endonuclease (TaKaRa, Dalian, Beijing, China). The standard strain *Salmonella enterica* serotype Braenderup H9812 digested with *Xba*I *was used as a marker. T*he electrophoresis was performed in 0.5 × TBE buffer in a pulsed-field electrophoresis system (Chef Mapper; Bio-Rad Laboratories, Hercules, CA, USA), and the conditions were as follows: 14°C, 6 V/cm, switch angle 120°, switch ramp 5–20 s for 19 h. BioNumerics software version 7.6 (Applied Maths, Sint-Martens-Latem, Belgium) was used to analyze the PFGE banding patterns. A cut off of 85% was used to judge the relatedness of strains analyzed based on the tree constructed by the unweighted pair group method of averages and a position tolerance of 1.5%.

Statistical analysis

IBM SPSS Statistics 20.0 software was used to perform statistical analysis. To determine whether there are statistical outliers among the fluorescence label clearance rates, we performed multivariate linear regression analysis to check their Mahalanobis distance. Different marker numbers in the 8 quarters was tested for Normal distribution. The correlation between the removal level of fluorescence labeling and colonization rates of MDR-AB, and the relationship between the colonization rates and infection rates of MDR-AB were analyzed by the Spearman correlation analysis. $P < 0.05$ was taken as statistically significant.

Results

Hand hygiene adherence in the comprehensive ICU

Totally, 676 opportunities for hand hygiene were obtained during 2013-2014 (Table 1). Clinicians contributed a majority of 51.18% of all opportunities followed by Nurse (35.95%) and Nursing assistants (12.87%). The overall adherence rates averaged about 61.76%. Adherence rates do vary by category of healthcare personnel, in general, compliance of the clinicians was the best, whereas, the adherence of environment service staff was poor. On the whole, hand hygiene compliance of healthcare personnel were relatively stable during 2013-2014, albeit hand hygiene compliance of clinicians increased from 68.6% to 76.6%, the adherence of hand hygiene of nurses and Nursing assistants showed a fluctuating trend

Clearance of fluorescence labeling

Considering that the numbers of fluorescent marks among the 8 quarters were different, fluctuating from 862 to 1199. Statistical analysis was conducted and showed that all of them conformed to the normal distribution (p=0.2).

At the initial stage, the clearance rate of fluorescence labeling was comparatively low, only 21.9% (Table 2). Through training and strengthening supervision of cleaning workers, the total clearance rates of fluorescence labeling were greatly improved and finally reached up to 85.7% at the last quarter of 2013. However, with the frequent change and mobility of cleaning workers within ICU during 2014, the average clearance rate sharply decreased to less than 50%, even though frequent straining and education were implemented.

The contamination rate of MDR-AB on the HTCS within ICUs

To monitor the contamination level of MDR-AB on the HTCS, samples were collected for bacterial culture and identification. The distribution of MDR-AB isolates on HTCS was displayed in Table 3. At the first quarter of 2013, 6 MDR-AB isolates were detected from HTCS, mainly on treatment vehicle and guardrail. With the increasing clearance rates of fluorescence labeling, the contamination rate of MDR-AB decreased remarkably, thus, in the following 3 quarters, no MDR-AB isolates were found. However, with the drastic fluctuation of fluorescence clearance rates during 2014, MDR-AB isolates were continuously detected from guardrail, treatment vehicle and treatment table.

The colonization and infection rates of MDR-AB among inpatient within the ICU

As shown in Table 4, in 2013, the hospital colonization rate of MDR-AB per Bed Days changed in the wake of the clearance level of fluorescence labeling. Spearman correlation analysis found a significant association between them (p=0.021). In addition, we found that the changes in the infection rates of MDR-AB were consistent with the ones in the colonization rates of inpatient within the ICU. However, we did not find a significant correlation between the colonization rates and the infection rates of MDR-AB.

Overall, the increased clearance rates of fluorescence marks lead to decreased contamination rates of MDR-AB on HTCS. And the colonization rates and infection rate of MDR-AB in inpatient also decreased correspondingly. Statistical analysis showed that the correlation equation between clear rates of fluorescence labeling HTCS and hospital infection rates per thousand bed days was , which indicated

that the latter decreased along with the former increase . It's worthy to mention that the clearance rate of January to March in 2013 (21.9%) was not statistical outlier, since its Mahalanobis distance was 5.31, which was less than threshold value of chi-square test (16.74) during multivariate linear regression analysis.

The genetic relatedness of the MDR-AB strains during the infection outbreak

From Jan to Mar in 2013, 7 MDR-AB strains were isolated from sputum samples of 7 inpatients. Among them, 3 strains were associated with HAIs (The positive sputum culture was drawn >2 days after admission) (28), 4 strains with community infections (the positive sputum culture was drawn <2 days after admission). At the same time, 6 MDR-AB strains were isolated from the HTCS of the 3 HAI patients (H01-H03). among them, 2 ones were isolated from treatment vehicle of Patient H01, 2 ones from guardrail and 1 one from the treatment vehicle of Patient H02, The last one from treatment table of Patient H03. According to the cutoff of 85%, all the MDR-AB displayed a genetic relatedness, indicating the existence of an epidemic clone (Figure 2).

Discussion

In this study, we utilized a fluorescence labeling method to systematically evaluate the cleanliness of environmental surfaces within a comprehensive ICU in Nanjing Drum Tower Hospital, a large tertiary hospital of Nanjing, southeast China. Furthermore, the effect of environmental cleaning and hand hygiene adherence on the colonization and infection rates of MDR-AB in patients was also investigated. We found that the cleanliness of environmental surfaces could be reflected by clearance rates of fluorescence labeling on HTCS, under the conditions of keeping the stability of environment serve staff and relatively stable hand hygiene compliance, the clearance rates of fluorescence labeling could be greatly improved by training and strengthening supervision of environmental services staff. In addition, increasing clearance rates of fluorescence labeling on HTCS was associated with the hospital infection rates of MDR-AB.

Hand hygiene and Environmental cleaning have previously been demonstrated to be two pillars of infection prevention in the control of hospital-acquired infection (29). Comparing with the progressively improved compliance from 48% to 66% in a teaching hospital in Switzerland (30) during a 3-year survey period, and the low adherence to hand hygiene of clinicians (30.7%) at Queen Elizabeth Central Hospital in Malawi (31), the overall compliance with hand hygiene in our study was relatively stable and high (61.76%), albeit there were higher rates of hand hygiene compliance in clinicians when compared to nurse and environmental services staff. Notably, we found a continuously increasing compliance of hand hygiene of clinicians, which suggests that more and more clinicians in our ICU have realized the importance of the hand hygiene in the control of nosocomial pathogen. Considering the relatively stable hand hygiene adherence, more attention was therefore payed to analyze the effect of environment cleaning produced by the removal of fluorescence labeling.

Compared with the 44% clearance rate of black-light marks at baseline on surfaces in ICUs in the United Kingdom (17), the quite low (21.9%) removal rate of fluorescence labeling in our study corresponded to a bad actual environmental cleaning, which means that more than 50% of the HTCS that should be wiped were actually not cleaned, indicating that cleaning of environmental surfaces should be strengthened. Fortunately, when the data were fed back to the management department of environmental services staff, the cleaning processes were optimized, due to more training and stricter supervision, the clearance rate of fluorescence labeling has been greatly increased to 85.7% in the last quarter of 2013, a little lower than the 94% removal rate of fluorescent marker in United kingdom (32), suggesting that education of environmental services staff, and feedback using fluorescence labeling monitoring system could greatly improve the thoroughness of environment cleaning. However, with the frequent change and mobility of cleaning workers in the year 2014, the environment cleaning reflected by constantly decreasing clearance rates of fluorescence labeling was greatly affected albeit regular training was still implemented, the importance of the stability of cleaning workers within ICUs is thus emphasized. Altogether, based on the fact that there are always much more patients and fewer beds in the large Third-Class A General Hospitals in China, it is difficult to achieve isolation measures strictly, such as single-room for each patient or the same-room placement for the same multidrug-resistant bacterial infections or colonizers. Thus, hand hygiene in combination with the formulated practical measures and training for all kinds of medical personnel to strengthen environmental cleaning as well as keeping the stability of environment service staff are very important to prevent transmission of drug-resistant bacteria. Additionally, fluorescence labeling is economical and effective method to rapidly and effectively evaluate the environmental clearance within ICUs.

Our further analysis found that there is a negative correlation between the clearance rates of fluorescence labeling on HTCS and the hospital infection rates per thousand bed days on the whole, which indicates that higher clearance rates of fluorescence labeling will produce the effect of lower hospital infection rates of MDR-AB within ICUs, providing evidence that fluorescence labeling is a rapid and effective method to monitor clearance of medical environment. Moreover, few MDR-AB colonization in inpatients corresponding to the gradually increasing clearance rates of fluorescence labeling on HTCS were observed during 2013, indicating that good environment cleaning on HTCS could result in decreases in patient colonization.

Furthermore, the close genetic relationship of the MDR-AB isolated from the environmental surfaces and specimens of inpatients suggested the existence of an epidemic MDR-AB, which could rapidly spread within ICUs once the environmental cleaning is not enough, further emphasizing the importance of the sanitation of hospital environment. Thus, strengthening the cleanness and disinfection of the HCTS and other infection prevention and control measures, including surveillance is an effective way to prevent and control the dissemination of MDR-AB within hospitals.

There are several limitations to this study. First, our study focused only on daily cleaning when the room is occupied not terminal cleaning after patient discharge. Second, we just cultured only a fraction of

marked surfaces because of financial constraints. Third, Anal swab were not taken for the surveillance for MDR-AB colonization.

Conclusion

Epidemic MDR-AB is a main pathogen easily colonizing on the HCTS within ICUs. with the fluorescence labeling method, the environmental cleanliness could be effectively reflected and educational intervention could also be objectively assessed. Furthermore, our study emphasized the importance of monitoring environment clearance and keeping stability environment serve staff under the condition of relatively stable hand hygiene compliance.

Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request

Competing interests

The authors declare that they have no competing interests

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Authors' contributions

Yang Li and Hai Ge implemented the fluorescent labeling, evaluation of fluorescent removal rates. Wanqing Zhou, Jie Zheng and Hui Zhou performed the bacterial identification, susceptibility testing, Wei Chen and Xiaoli Cao analyzed the data and revision on the manuscripts.

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References

1. Ling ML, Apisarnthanarak A, Madriaga G. The Burden of Healthcare-Associated Infections in Southeast Asia: A Systematic Literature Review and Meta-analysis. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2015;60(11):1690-9.
2. Alvarez A, Fernandez L, Gutierrez D, Iglesias B, Rodriguez A. Methicillin-Resistant Staphylococcus aureus in Hospitals: Latest Trends and Treatments Based on Bacteriophages. J Clin Microbiol. 2019;57(12): e01006-19..
3. Milstone AM, Song X, Beers C, Berkowitz I, Carroll KC, Perl TM. Unrecognized burden of methicillin-resistant Staphylococcus aureus and vancomycin-resistant Enterococcus carriage in the pediatric intensive care unit. Infection control and hospital epidemiology. 2008;29(12):1174-6.
4. Kang J, Sickbert-Bennett EE, Brown VM, Weber DJ, Rutala WA. Changes in the incidence of health care-associated pathogens at a university hospital from 2005 to 2011. American journal of infection control. 2014;42(7):770-5.
5. Ramm L, Siani H, Wesgate R, Maillard JY. Pathogen transfer and high variability in pathogen removal by detergent wipes. American journal of infection control. 2015;43(7):724-8.
6. Zhanel GG, Decorby M, Nichol KA, Baudry PJ, Karlowsky JA, Lagace-Wiens PR, et al. Characterization of methicillin-resistant Staphylococcus aureus, vancomycin-resistant enterococci and extended-spectrum beta-lactamase-producing Escherichia coli in intensive care units in Canada: Results of the Canadian National Intensive Care Unit (CAN-ICU) study (2005-2006). The Canadian journal of infectious diseases & medical microbiology = Journal canadien des maladies infectieuses et de la microbiologie medicale. 2008;19(3):243-9.
7. Lazaris A, Coleman DC, Kearns AM, Pichon B, Kinnevey PM, Earls MR, et al. Novel multiresistance cfr plasmids in linezolid-resistant methicillin-resistant Staphylococcus epidermidis and vancomycin-resistant Enterococcus faecium (VRE) from a hospital outbreak: co-location of cfr and optrA in VRE. The Journal of antimicrobial chemotherapy. 2017;72(12):3252-7.
8. Zhang J, Zhao C, Chen H, Li H, Wang Q, Wang Z, et al. A multicenter epidemiology study on the risk factors and clinical outcomes of nosocomial intra-abdominal infections in China: results from the Chinese Antimicrobial Resistance Surveillance of Nosocomial Infections (CARES) 2007-2016. Infection and drug resistance. 2018;11:2311-9.
9. Sui W, Wang J, Wang H, Wang M, Huang Y, Zhuo J, et al. Comparing the transmission potential of Methicillin-resistant Staphylococcus aureus and multidrug-resistant Acinetobacter baumannii among

inpatients using target environmental monitoring. American journal of infection control. 2013;41(5):411-5.

10. Huang X, Li G, Yi L, Li M, Wang J. [The epidemiology of multidrug-resistant bacteria colonization and analysis of its risk factors in intensive care unit]. Zhonghua wei zhong bing ji jiu yi xue. 2015;27(8):667-71.
11. Zhao Y, Hu K, Zhang J, Guo Y, Fan X, Wang Y, et al. Outbreak of carbapenem-resistant Acinetobacter baumannii carrying the carbapenemase OXA-23 in ICU of the eastern Heilongjiang Province, China. 2019;19(1):452.
12. Haverkate MR, Derde LP, Brun-Buisson C, Bonten MJ, Bootsma MC. Duration of colonization with antimicrobial-resistant bacteria after ICU discharge. Intensive care medicine. 2014;40(4):564-71.
13. Liu WJ, Fu L, Huang M, Zhang JP, Wu Y, Zhou YS, et al. Frequency of antiseptic resistance genes and reduced susceptibility to biocides in carbapenem-resistant Acinetobacter baumannii. Journal of medical microbiology. 2017;66(1):13-7.
14. Nasr P. Genetics, epidemiology, and clinical manifestations of multidrug-resistant Acinetobacter baumannii. The Journal of hospital infection. 2020;104(1):4-11. doi: 10.1016/j.jhin.2019.09.021.
15. Sehulster L, Chinn RY. Guidelines for environmental infection control in health-care facilities. Recommendations of CDC and the Healthcare Infection Control Practices Advisory Committee (HICPAC). MMWR Recomm Rep. 2003;52(Rr-10):1-42.
16. Carling PC, Briggs J, Hylander D, Perkins J. An evaluation of patient area cleaning in 3 hospitals using a novel targeting methodology. American journal of infection control. 2006;34(8):513-9.
17. Goodman ER, Platt R, Bass R, Onderdonk AB, Yokoe DS, Huang SS. Impact of an environmental cleaning intervention on the presence of methicillin-resistant Staphylococcus aureus and vancomycin-resistant enterococci on surfaces in intensive care unit rooms. Infect Control Hosp Epidemiol. 2008;29(7):593-9.
18. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske CG, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases. 2012;18(3):268-81.
19. WHO Guidelines Approved by the Guidelines Review Committee. WHO Guidelines on Hand Hygiene in Health Care: First Global Patient Safety Challenge Clean Care Is Safer Care. Geneva: World Health Organization Copyright © 2009, World Health Organization.; 2009.
20. Boyce JM. Hand hygiene compliance monitoring: current perspectives from the USA. J Hosp Infect. 2008;70 Suppl 1:2-7.
21. Albert RK, Condie F. Hand-washing patterns in medical intensive-care units. N Engl J Med. 1981;304(24):1465-6.

22. Pittet D, Mourouga P, Perneger TV. Compliance with handwashing in a teaching hospital. Infection Control Program. Ann Intern Med. 1999;130(2):126-30.
23. Katz JD. Hand washing and hand disinfection: more than your mother taught you. Anesthesiol Clin North Am. 2004;22(3):457-71, vi.
24. Carling PC, Von Beheren S, Kim P, Woods C. Intensive care unit environmental cleaning: an evaluation in sixteen hospitals using a novel assessment tool. The Journal of hospital infection. 2008;68(1):39-44.
25. Horan TC, Andrus M, Dudeck MA. CDC/NHSN surveillance definition of health care-associated infection and criteria for specific types of infections in the acute care setting. American journal of infection control. 2008;36(5):309-32.
26. CLSI. Performance Standards for Antimicrobial Susceptibility Testing. . Clinical Laboratory Standard Institute. 2015; M100-S25.
27. Alcantar-Curiel MD, Rosales-Reyes R, Jarillo-Quijada MD, Gayosso-Vazquez C, Fernandez-Vazquez JL, Toledano-Tableros JE, et al. Carbapenem-Resistant Acinetobacter baumannii in Three Tertiary Care Hospitals in Mexico: Virulence Profiles, Innate Immune Response and Clonal Dissemination. Frontiers in microbiology. 2019;10:2116.
28. Buetti N, Atkinson A, Kronenberg A, Marschall J. Different Epidemiology of Hospital-Acquired Bloodstream Infections Between Small Community Hospitals and Large Community Hospitals. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2017;64(7):984-5.
29. Barnes SL, Morgan DJ, Harris AD, Carling PC, Thom KA. Preventing the transmission of multidrug-resistant organisms: modeling the relative importance of hand hygiene and environmental cleaning interventions. Infect Control Hosp Epidemiol. 2014;35(9):1156-62.
30. Pittet D, Hugonnet S, Harbarth S, Mourouga P, Sauvan V, Touveneau S, et al. Effectiveness of a hospital-wide programme to improve compliance with hand hygiene. Infection Control Programme. Lancet. 2000;356(9238):1307-12.
31. Kalata NL, Kamange L, Muula AS. Adherence to hand hygiene protocol by clinicians and medical students at Queen Elizabeth Central Hospital, Blantyre-Malawi. Malawi Med J. 2013;25(2):50-2.
32. Carling PC, Briggs JL, Perkins J, Highlander D. Improved cleaning of patient rooms using a new targeting method. Clin Infect Dis. 2006;42(3):385-8.

Tables

Table 1. Hand hygiene compliance

Medical staff		Time period							
		1-3, 2013	4-6, 2013	7-9, 2013	10-12, 2013	1-3, 2014	4-6, 2014	7-9, 2014	10-12, 2014
Clinicians	Number of opportunities	51	34	43	43	42	48	38	47
	Number of executions	35	24	31	30	32	34	28	36
	Compliance rate	68.63	70.59	72.09	69.77	76.19	70.83	73.68	76.60
Nurse	Number of opportunities	42	25	38	19	29	29	35	26
	Number of executions	24	14	18	9	19	16	20	18
	Compliance rate	57.14	56.00	47.37	47.37	65.52	55.17	57.14	69.23
Environment service staff	Number of opportunities	9	7	18	3	8	15	12	15
	Number of executions	4	3	6	2	2	4	0	7
	Compliance rate	44.44	42.86	33.33	66.67	25.00	26.67	0	46.67
Total	Number of opportunities	102	66	99	65	79	92	85	88
	Number of executions	63	41	55	41	53	54	48	61
	Compliance rate	61.76	62.12	55.56	63.08	67.09	58.70	56.47	69.32

Table 2. The number of fluorescent marks on high frequency contact sites and clearance rates of fluorescence labeling

HTCS	The number of fluorescent markers and removal	Time period							
		1-3, 2013	4-6, 2013	7-9, 2013	10-12, 2013	1-3, 2014	4-6, 2014	7-9, 2014	10-12, 2014
Guardrail	Marker number	520	431	463	531	568	446	420	467
	Scavenging number	102	139	377	489	283	186	175	173
Bedside table	Marker number	116	124	112	127	108	91	74	85
	Scavenging number	23	89	100	115	75	47	44	31
Treatment vehicle	Marker number	132	97	144	105	87	96	111	93
	Scavenging number	54	73	124	87	39	39	57	55
Monitor button	Marker number	194	108	137	99	115	127	100	86
	Scavenging number	31	43	87	76	47	53	41	26
Injection pump button	Marker number	191	84	154	116	152	110	242	131
	Scavenging number	58	27	119	84	70	31	73	48
Treatment table	Marker number	46	18	33	29	19	15	41	62
	Scavenging number	1	7	28	12	5	6	17	21
total	Marker number	1199	862	1043	1007	1049	885	988	924
	Scavenging number	269	378	835	863	519	362	407	354
Clearance rate (%)		21.9	43.9	80	85.7	49.5	40.9	41.2	38.3

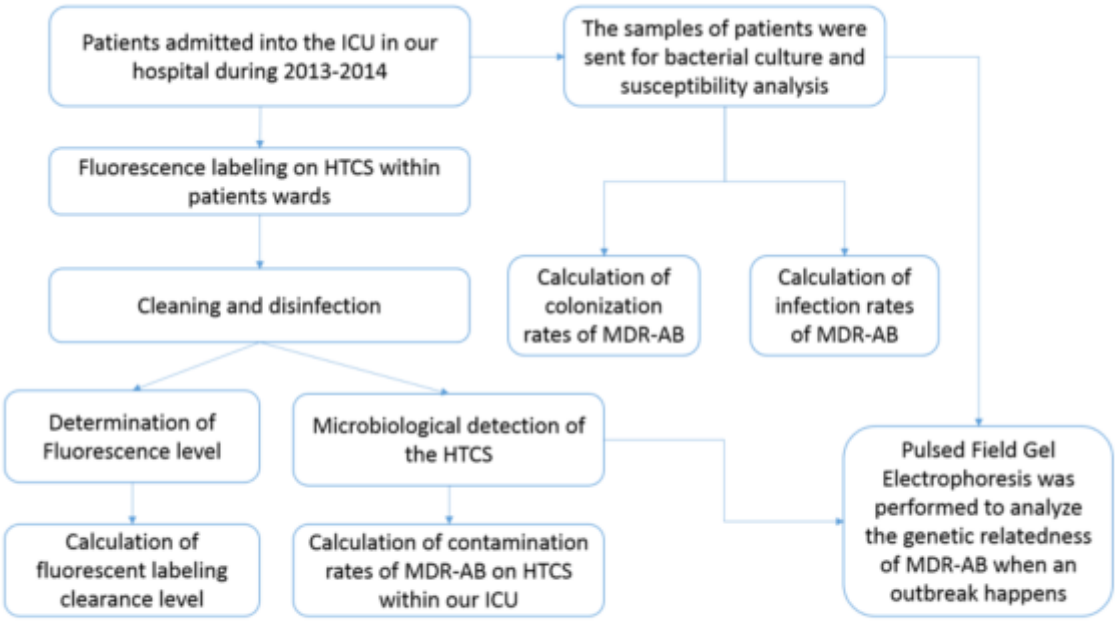
Table 3. Distribution of multidrug-resistant *Acinetobacter baumannii* on HTCS

HTCS	The number of fluorescent markers and removal	Time period							
		1-3, 2013	4-6, 2013	7-9, 2013	10-12, 2013	1-3, 2014	4-6, 2014	7-9, 2014	10-12, 2014
Guardrail	Number of samples	10	10	10	8	8	8	8	8
	Number of positive samples	2	0	0	0	1	0	1	1
Bedside table	Number of samples	6	5	4	3	3	3	4	4
	Number of positive samples	0	0	0	0	0	0	0	0
Treatment vehicle	Number of samples	8	10	8	6	4	4	6	8
	Number of positive samples	3	0	0	0	0	0	1	1
Monitor button	Number of samples	3	2	3	3	4	4	3	3
	Number of positive samples	0	0	0	0	0	0	0	0
Injection pump button	Number of samples	3	2	3	3	4	4	3	3
	Number of positive samples	0	0	0	0	0	0	0	0
Treatment table	Number of samples	3	3	3	3	4	5	6	6
	Number of positive samples	1	0	0	0	0	1	1	0
total	Number of samples	33	32	31	26	27	28	30	32
	Number of positive samples	6	0	0	0	1	1	3	2
Clearance rate (%)		21.9	43.9	80	85.7	49.5	40.9	41.2	38.3

Table 4. The clearance rates of fluorescent marks, the colonization rates of multidrug-resistant *Acinetobacter baumannii* and the infection rates of multidrug-resistant *Acinetobacter baumannii* of the inpatients within our ICU

Time period	Fluorescent label clearance rates (%)	Bed Days	The number of MDR-AB for colonization of inpatients	The number of MDR-AB for healthcare-associated infections	The daily colonization rates of MRD-AB per thousand bed (‰)	The hospital infection rates of MDR-AB per thousand bed days (‰)
1-3, 2013	21.9	1761	29	13	16.47	7.38
4-6, 2013	43.9	1845	33	3	17.89	1.63
7-9, 2013	80	1542	14	7	9.08	4.54
10-12, 2013	85.7	1775	10	5	5.63	2.82
1-3, 2014	49.5	1972	18	3	9.13	1.52
4-6, 2014	40.9	1824	30	12	16.45	6.58
7-9, 2014	41.2	1779	15	5	8.43	2.81
10-12, 2014	38.3	1810	20	7	11.05	3.87

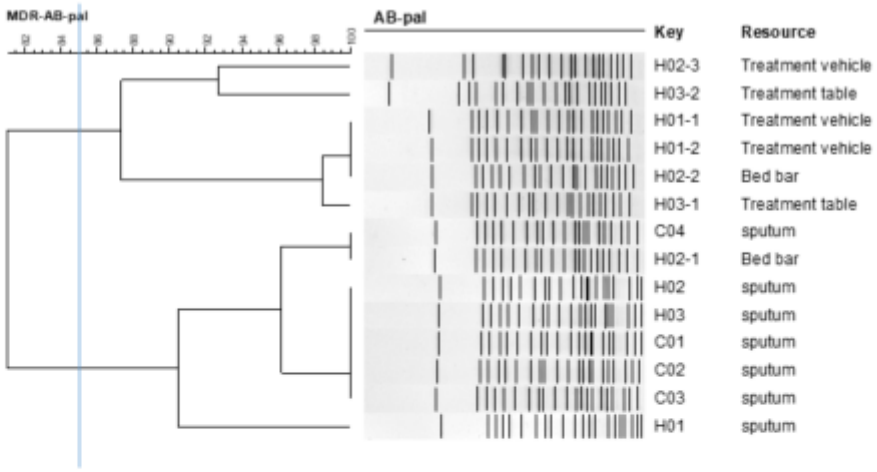
Figures



ICU: Intensive Care Unit; MDR-AB: multidrug resistant *Acinetobacter baummannii*; HTCS, high-touch clinical surfaces.

Figure 1

ICU: Intensive Care Unit; MDR-AB: multidrug resistant Acinetobacter baummannii; HTCS, high-touch clinical surfaces.



Dendrogram based on PFGE profiles of 12 multidrug-resistant *Acinetobacter baumannii* isolated from inpatients and environment surfaces within ICU. The dendrogram was produced by the UPGMA algorithm based on the Dice similarity coefficient. H01, H02 and H03 were associated with hospital acquired infections; C01, C02, C03 and C04 were associated with community acquired infections; H01-1, H01-2 were isolated from the HTCS of H01 infected patient; H02-1, H02-2, H02-3 from the HTCS of H02 infected patient; H03-1 and H03-2 from the HTCS of H03 infected patient;

Figure 2

Dendrogram based on PFGE profiles of 12 multidrug-resistant Acinetobacter baumannii isolated from inpatients and environment surfaces within ICU. The dendrogram was produced by the UPGMA algorithm based on the Dice similarity coefficient. H01, H02 and H03 were associated with hospital acquired infections; C01, C02, C03 and C04 were associated with community acquired infections; H01-1, H01-2 were isolated from the HTCS of H01 infected patient; H02-1, H02-2, H02-3 from the HTCS of H02 infected patient; H03-1 and H03-2 from the HTCS of H03 infected patient;