Infection control for a methicillin-resistant *Staphylococcus aureus* outbreak in an advanced emergency medical service center, as monitored by molecular analysis

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Running title: *Infection Control for MRSA Outbreak*

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SUMMARY

Background: A methicillin-resistant *Staphylococcus aureus* (MRSA) outbreak occurred in an advanced emergency medical service center between 2010 and 2011.

Aim: Our objective was to evaluate the status of the MRSA outbreak, as monitored by molecular analysis.

Methods: Twenty-eight MRSA strains were isolated from the blood of 11 patients, and from other specimens (pharynx, nasal cavity, etc) of 12 patients, the environments, and the skin, middle nutus and urine of each one patient from other wards. Pulsed-field gel electrophoresis (PFGE) was performed to evaluate horizontal transmission.

Results: Molecular typing by PFGE showed the 28 MRSA strains had 7 patterns, and the PFGE patterns of the 11 MRSA strains were identical. Unselective use of intranasal mupirocin ointment, MRSA monitoring for new inpatients and the prevention of direct or indirect contact infection were performed. However, the number of inpatients with MRSA did not quickly decreased, and additional molecular typing by PFGE showed 10 of 19 MRSA strains (5 of 6 from blood, 5 of 13 from other specimens) were still identical. Lectures and ward rounds were performed repeatedly, and participation in ward rounds by staffs was suggested. Finally, the number of inpatients with MRSA was significantly decreased more than 6 months after the intervention. Although the MRSA outbreak was thought to have ended, follow-up molecular typing by PFGE showed horizontal transmission persisted.

Conclusion: our data suggest that various and combined measures of infection control are essential and monitoring by molecular analysis using PFGE is useful to identify the status of a MRSA outbreak.

INTRODUCTION

Methicillin-resistant *Staphylococcus aureus*

(MRSA) is a major causative organism of hospital-acquired infection. MRSA strains easily colonize a host, particularly immunodeficient patients, and can cause a variety of serious infections [1-3]. The principal mode of transmission is via the transiently colonized hands of hospital personnel [4]. An outbreak of MRSA in intensive care units or neonatal intensive care units is often prolonged and can result in substantial morbidity and mortality [5, 6]. Although there are reports concerning the efficacy of unselective use of intranasal mupirocin ointment to control MRSA outbreak [7, 8], effective infection control measures still have not been established.

Molecular analysis is essential for the evaluation of horizontal transmission during a MRSA outbreak, and pulsed-field gel electrophoresis (PFGE) remains to be the gold standard [9-11].

We describe the control of a MRSA outbreak in our advanced emergency medical service center (hereafter referred to as the ICU) through early recognition of the outbreak, monitoring by molecular analysis and the stepwise addition of infection control measures.

METHODS

Ethical Approval

All studies described herein had been approved by the Human Ethics Review Boards of Kurume University (10268).

Setting and Outbreak description

In Kurume university hospital, there are 29 diagnosis and treatment departments, and 25 wards with 1,098 beds including an ICU with 44 beds. The number of monthly patients with newly identified MRSA colonization or infections including a blood-culture positive in the ICU had increased beginning in June, 2010 (Figure 1). Since an outbreak is defined as at least twice the usual number of cases in the same ward, the Infection Control Team (ICT) started the intervention for a MRSA outbreak in the ICU beginning in August, 2010.

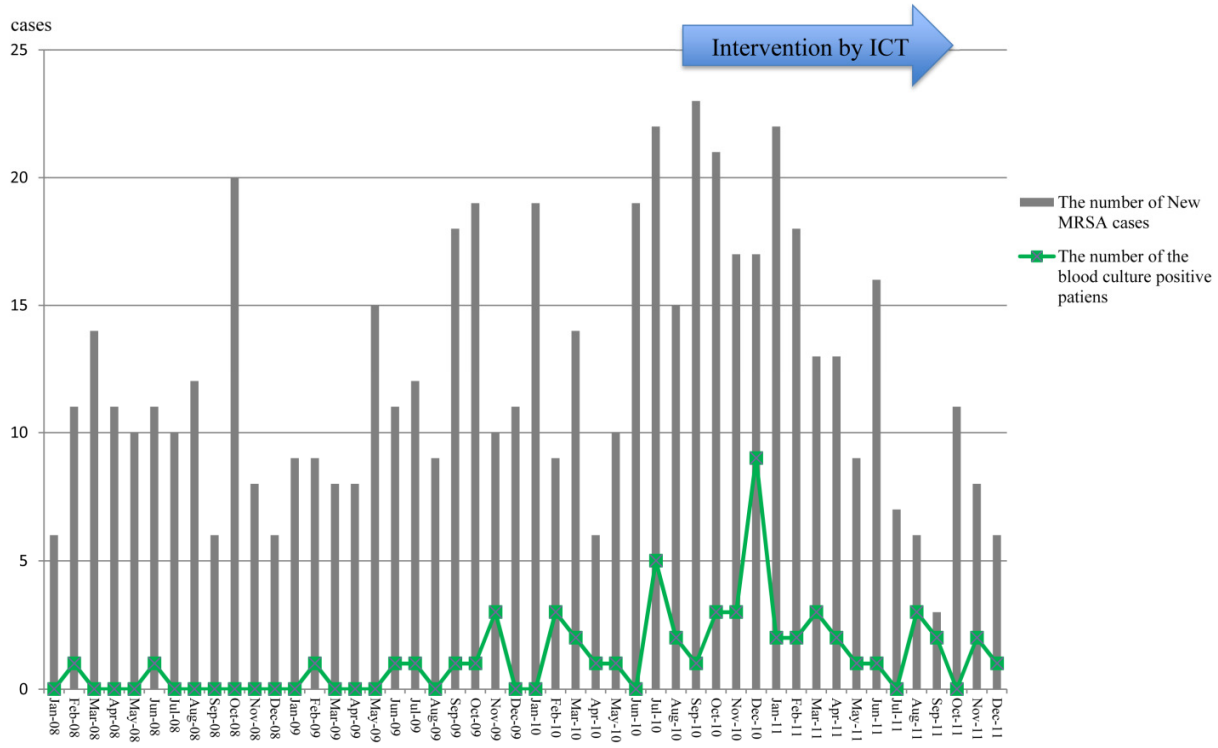


Figure 1. Transition of monthly patients with newly identified MRSA colonization or infection including blood culture positive in the ICU from January, 2008 to December, 2011. MRSA cases with blood culture positive patients in the ICU dramatically increased from June, 2010. As a result of having performed various kinds of interventions, the occurrence of MRSA infections was decreased more than 6 months after intervention was begun.

Figure 2. Post-intervention improvements by the ICT. Left: Organizing a desk for injection manufacture. Center: Clarification of waste containers. Right: Placement of a rack for gloves and gowns.

Bacterial strains and patients

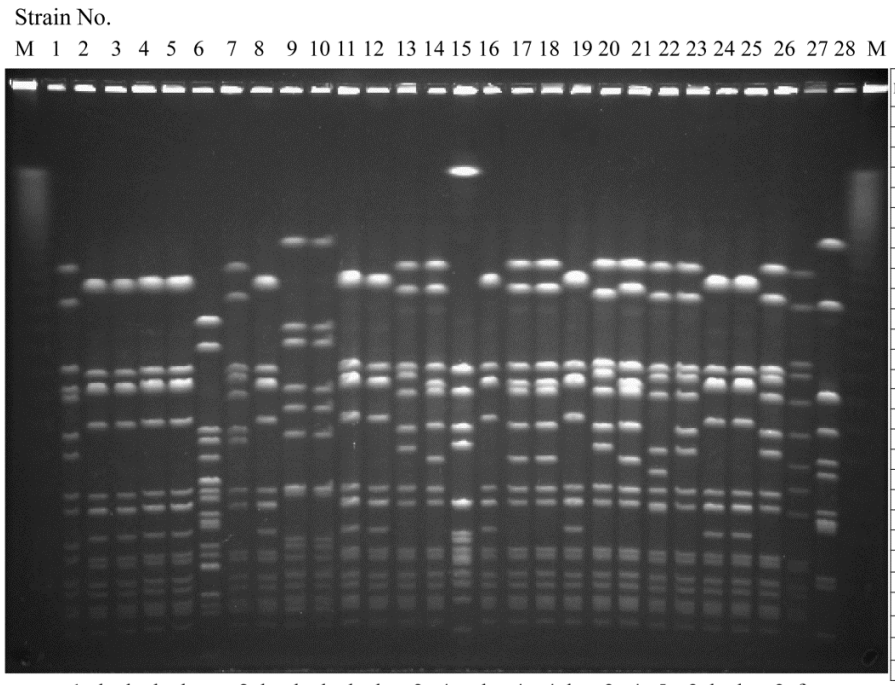
The first PFGE was performed against 28 MRSA isolates from the blood (11 strains) of 11 patients, and from other specimens (pharynx, nasal cavity, stool, sputum, ascites and pus) of 12 patient (12 strains), the environments (2 strains), and the skin, middle nutus and urine of each one patient (3 strains) from other wards between April and September, 2010. Since the number of inpatients with MRSA had not decreased quickly despite various infection control measures such as unselective use of intranasal mupirocin ointment to inpatients, MRSA monitoring for new inpatients and measures for the prevention of direct or indirect contact were performed, A second PFGE was performed against 19 MRSA isolates from blood, sputum, nasal cavity, skin and pus in the ICU, and each of 3 MRSA strains in neurosurgery, reconstructive and maxillofacial surgery and cardiovasucular medicine where inpatients were occasionally transferred from the ICU between October, 2010 and January, 2011. Finally, even after the end of the MRSA outbreak in the ICU a follow-up PFGE was performed against 28 MRSA isolates from blood, nasal cavities, sputum, pus, pharynx, catheter tip and ascites in the ICU between August and November, 2011.

Pulsed-field gel electrophoresis

PFGE was performed as described previously [1]. The DNA were digested with SmaI (Takara Shuzo Co., Shiga, Japan). CHEF Mapper Pulsed Field Electrophoresis Systems (Bio-Rad Life Science Group, Hercules, CA, USA) were used for the electrophoresis, with a potential of 6 V/cm, switch times of 0.47 and 63 seconds, and a run-time of 20 hours and 18 minutes. After staining with ethidium bromide, the interpretation of PFGE patterns was based on the criteria described by Tenover *et al* [12].

Intervention by the ICT

A weekly MRSA monitoring culture for all inpatients in the ICU was continued from August, 2010, and environmental bacterial culture in 38 locations in the ICU were done in September, 2010. MRSA was detected in 2 of the 38 locations (5.3%) around MRSA colonized patients. As molecular analysis by the first PFGE



No.	Date (mo/day/yr)	Source	Location
1	04.12.10	Blood	ICU
2	05.10.10	Blood	ICU
3	06.16.10	Blood	ICU
4	06.18.10	Blood	ICU
5	06.22.10	Blood	ICU
6	06.22.10	Blood	ICU
7	06.24.10	Blood	ICU
8	07.20.10	Blood	ICU
9	07.26.10	Blood	ICU
10	08.10.10	Blood	ICU
11	09.21.10	Blood	ICU
12	08.25.10	Pharynx	ICU
13	08.28.10	Stool	ICU
14	08.30.10	Pharynx	ICU
15	08.31.10	Pharynx	ICU
16	08.31.10	Pus	ICU
17	08.31.10	Pharynx	ICU
18	08.31.10	Nasal cavity	ICU
19	09.01.10	Ascites	ICU
20	09.04.10	Nasal cavity	ICU
21	09.07.10	Nasal cavity	ICU
22	09.08.10	Stool	ICU
23	09.14.10	Sputum	ICU
24	09.10.10	Suction tube	ICU
25	09.10.10	Ventilator panel	ICU
26	08.30.10	Skin	Dermatology
27	09.02.10	Middle nutus	Otolaryngology
28	09.02.10	Urine	Pediatrics

ICU: advanced emergency medical service center

Figure 3. PFGE patterns of Sma I digested DNAs of MRSA isolates between April and September, 2010. Molecular typing of the first PFGE showed 28 MRSA strains had 7 patterns, and the PFGE patterns of 11 MRSA strains (blood in 6, environment in 2, and pharynx, ascites and pus in each one) were identical、and the predominant PFGE pattern b was not confirmed in other wards (No. 26-28).

showed the horizontal transmission of MRSA strains in the ICU, the ICT began the following measures: 1) active surveillance and unselective use of intranasal mupirocin ointment to all inpatients for 3 days beginning in November, 2010, 2) reinforcement of direct or indirect contact infection measures, 3) Reinforcement of blood stream infection measures, and 4) action to monitor reinforcement by ICT and education for the ICU staffs (Figure 2).

RESULTS

Interpretation of PFGE

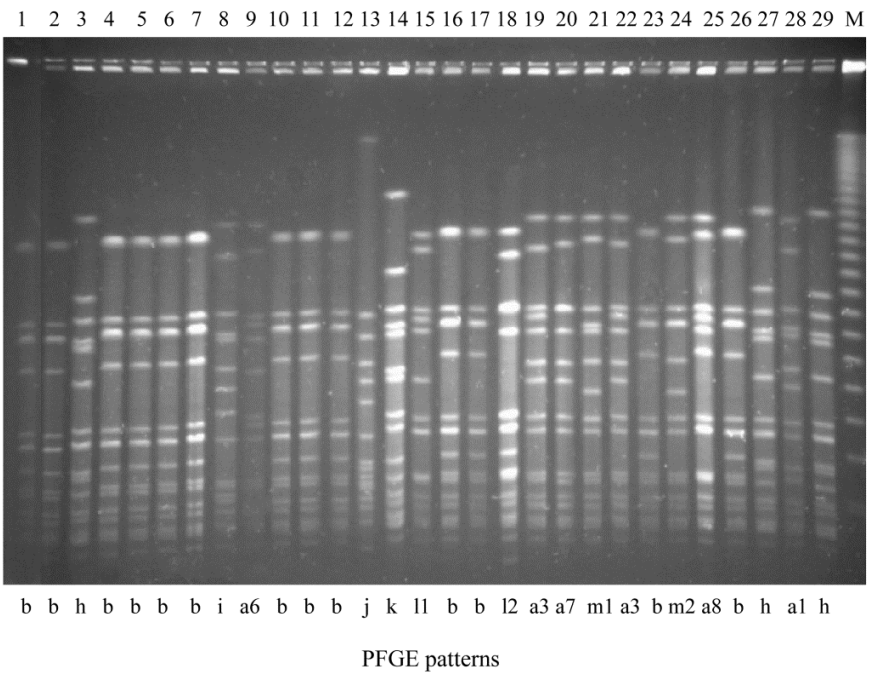
Molecular typing by the first PFGE showed 28 MRSA strains had 7 patterns, and the PFGE pattern b of 11 MRSA strains (blood in 6, environment in 2, and pharynx, ascites and pus in one each) were identical, which meant that the horizontal transmission of a predominant PFGE pattern b of the MRSA strains had occurred in the ICU (Figure 3).

Molecular typing by a second PFGE which was performed during the MRSA outbreak showed 19 MRSA strains in the ICU had 7 patterns, and 10 of the 19 MRSA strains (5 of 6 in blood, 5 of 13 in other specimens) were still the predominant PFGE pattern b (Figure 4). Moreover, the predominant PFGE pattern b of MRSA strain was confirmed in the patients who were transferred from the ICU to neurosurgery and reconstructive and maxillofacial surgery ward, which meant that the MRSA outbreak had potentially spread from the ICU to other wards.

Molecular typing by the follow-up PFGE even after the end of the MRSA outbreak in the ICU showed 28 MRSA strains had 7 patterns, and that 21 of 28 MRSA strains (2 of 4 in blood, 19 of 24 in other specimens) were still the PFGE pattern b (Figure 5), which meant that the horizontal transmission of the predominant PFGE pattern b of MRSA strains had continued even after the end of the MRSA outbreak in the ICU.

Additional intervention by ICT

The ICT performed various kinds of intervention for the prevention of MRSA outbreak in the ICU, as described above. However, the



No.	Date (mo/day/yr)	Source	Location
1	05.10.10	Blood	ICU
2	12.09.10	Blood	ICU
3	12.11.10	Blood	ICU
4	12.27.10	Blood	ICU
5	01.07.11	Blood	ICU
6	01.17.11	Blood	ICU
7	01.17.11	Blood	ICU
8	01.02.11	Sputum	ICU
9	01.04.11	Sputum	ICU
10	01.04.11	Nasal cavity	ICU
11	01.04.11	Skin	ICU
12	01.05.11	Pus	ICU
13	01.06.11	Sputum	ICU
14	01.11.11	Nasal cavity	ICU
15	01.11.11	Nasal cavity	ICU
16	01.12.11	Sputum	ICU
17	01.12.11	Sputum	ICU
18	01.12.11	Nasal cavity	ICU
19	01.17.11	Nasal cavity	ICU
20	01.17.11	Pus	ICU
21	11.01.10	Sputum	Neurosurgery
22	11.05.10	Pus	Neurosurgery
23	11.07.10	Spinal fluid	Neurosurgery
24	10.22.10	Pharynx	Reconstructive and Maxillofacial Surgery
25	11.01.10	Sputum	Reconstructive and Maxillofacial Surgery
26	11.10.10	Pus	Reconstructive and Maxillofacial Surgery
27	10.21.10	Pus	Cardiovascular Medicne
28	11.01.10	Sputum	Cardiovascular Medicne
29	11.04.10	Pharynx	Cardiovascular Medicne

ICU: advanced emergency medical service center

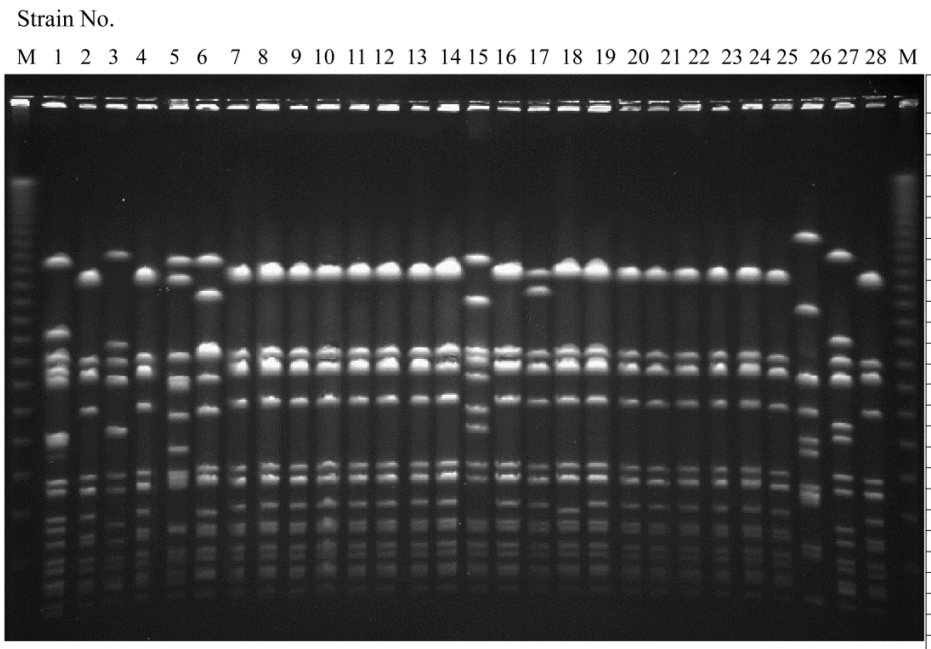
Figure 4. PFGE patterns of Sma I digested DNAs of MRSA isolates between October, 2010 and January, 2011. Molecular typing by a second PFGE showed 19 MRSA strains in the ICU had 7 patterns, and 10 of the 19 MRSA strains (5 of 6 in blood, 5 of 13 in other specimens) continued to be the predominant PFGE pattern b. The predominant PFGE pattern b of MRSA strains was confirmed in the patients who were transferred from the ICU to neurosurgery and reconstructive and to maxillofacial surgery wards (No. 23 and 26).

number of inpatients with MRSA did not quickly decrease. Lectures and ward rounds were repeatedly done, and participation in the ward rounds by the ICU staffs members was suggested to enhance the precaution against the MRSA outbreak. As a result of having performed various kinds of intervention, the number of inpatients with MRSA was significantly decreased more than 6 months later (Figure1).

DISCUSSION

MRSA outbreak occasionally occur as the result of hospital acquired infection in a hospital, particularly in intensive care units [13], neonatal intensive care unit [6] or burns unit [14], and often prolong and increase the incidences of hospital acquired pneumonia, blood stream infection and the risk of death [5]. Since the MRSA strain is known to spread in a ward via the transiently colonized hands of hospital personel [4], hand hygiene is a basic preventive measure [15]. Although a previous report demonstrated that simple control measures including contact isolation of colonized patients and reinforcement of hand washing practices among personnel has effectively controlled MRSA outbreaks [13], some specific preventive measure such as unselective use of intranasal mupirocin ointment [7, 8], and enteral vancomycin [16, 17] has eventually been used in clinical settings in the past. Indeed, various kinds of interventions including the unselective use of intranasal mupirocin ointment to control MRSA outbreaks in the ICU have been performed. However, the number of inpatients with MRSA was not decreased quickly, and the MRSA outbreak ended after additional interventions such as repeated lectures and ward rounds, and participation of ward rounds by the ICU staffs to enhance their precautions against further MRSA outbreaks.

During this particular MRSA outbreak, molecular analysis by PFGE was repeatedly performed to evaluate the horizontal transmission



No.	Date (mo/day/yr)	Source	Location
1	08.18.11	Blood	ICU
2	10.10.11	Blood	ICU
3	10.13.11	Blood	ICU
4	11.04.11	Blood	ICU
5	08.16.11	Nasal cavity	ICU
6	08.17.11	Sputum	ICU
7	08.19.11	Sputum	ICU
8	08.19.11	Sputum	ICU
9	08.20.11	Pus	ICU
10	08.29.11	Sputum	ICU
11	09.09.11	Sputum	ICU
12	09.12.11	Pharynx	ICU
13	09.27.11	Catheter tip	ICU
14	09.28.11	Pus	ICU
15	10.05.11	Ascites	ICU
16	10.03.11	Sputum	ICU
17	10.11.11	Sputum	ICU
18	10.11.11	Sputum	ICU
19	10.12.11	Sputum	ICU
20	10.13.11	Sputum	ICU
21	10.15.11	Sputum	ICU
22	10.17.11	Sputum	ICU
23	10.21.11	Ascites	ICU
24	10.31.11	Pus	ICU
25	11.07.11	Sputum	ICU
26	11.21.11	Sputum	ICU
27	11.21.11	Sputum	ICU
28	11.21.11	Sputum	ICU

ICU: advanced emergency medical service center

Figure 5. PFGE patterns of Sma I digested DNAs of MRSA isolates between August and November, 2011. Molecular typing by follow-up PFGE even after the end of MRSA outbreak in ICU showed 28 MRSA strains had 7 patterns, 21 of 28 MRSA strains (2 of 4 in blood, 19 of 24 in other specimens) were still predominant PFGE pattern b.

and the ICU staffs were immediately informed of the results. PFGE seems to be useful in evaluating the presence of cross-transmission in hospital- acquired infection [9-11]. In addition, previous study indicates that PFGE studies may be helpful in assessing the effectiveness of interventions during a MRSA outbreak [18]. In our study, a predominant MRSA isolate was thought to be spread in the ICU via the transiently colonized hands of hospital personnel. However, the follow-up PFGE even after the end of the MRSA outbreak in the ICU showed that the predominant MRSA isolate continued to spread as a horizontal transmission, although the number of inpatients with newly identified MRSA colonization or infections had decreased. That means that the predominant MRSA strain could potentially cause another MRSA outbreak in the ICU, and, therefore, the precautions should be continued.

In conclusion, our data suggest that various and combined measures of infection control are essential against a MRSA outbreak and that monitoring by molecular analysis using PFGE is useful in identifying the status of a MRSA outbreak. Therefore, the use of follow-up PFGE—even after the end of a MRSA outbreak—should be considered in order to prevent a recurrence.

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CONFLICT OF INTEREST

The authors declare that they have no conflict and interest.

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