Comprehensive Analysis of Systemically Disseminated ST8/non-USA300 type Community-acquired Methicillin-resistant Staphylococcus aureus Infection

Hideharu Hagiya¹,², Junzo Hisatsune³,⁴, Taro Kojima³,⁴, Sumiko Shiota⁵, Hiromichi Naito³, Shingo Hagioka², Naoki Morimoto⁷, Fumio Otsuka¹ and Motoyuki Sugai³,⁴

Abstract

Community-acquired methicillin-resistant Staphylococcus aureus (CA-MRSA) is genetically heterogeneous and various genotypes are spreading worldwide. We herein report a case of systematically disseminated Japan-intrinsic CA-MRSA infection that was successfully treated. A genetically identical single strain was isolated from a total of 11 different parts of the patient’s body, and the pathogen was found to be multilocus sequence type 8, staphylococcal cassette chromosome mec IV, coagulase type III and negative for both Panton-Valentine leukocidin and arginine catabolic mobile element. The epidemiology and pathogenicity of the Japan-intrinsic CA-MRSA strain remain unknown, and further investigation of this life-threatening organism is warranted.

Key words: community-acquired methicillin-resistant Staphylococcus aureus, multilocus sequence type 8, sepsis, toxic shock syndrome, USA300

(Intern Med 53: 907-912, 2014)
(DOI: 10.2169/internalmedicine.53.1746)

Introduction

Methicillin-resistant Staphylococcus aureus (MRSA) was first reported in the United Kingdom in 1961 (1). Since the 1990s, community-acquired methicillin-resistant Staphylococcus aureus (CA-MRSA) has newly emerged as a life-threatening pathogen worldwide (2). CA-MRSA has the ability to cause unusual severe infections, such as sepsis, necrotizing pneumonia, skin and soft tissue infection and osteomyelitis, even in healthy individuals without particular risk factors (3, 4), and is now becoming a leading highly virulent pathogen in communities (5, 6). Limited data are available regarding the morbidity and mortality of this pathogen; however, the mortality rate of CA-MRSA infection has been reported to be relatively high compared to that of healthcare-associated MRSA (7).

MRSA is genetically heterogeneous, and various genotypes are currently spreading worldwide (5). Although multilocus sequence type (ST) 1 was once dominant, the worldwide dominant types at present are reported to be ST8, 30, 59 and 80 (8-11). Among them, ST8, particularly the USA300 strain, has been reported to be the most highly virulent and prevalent type and is frequently isolated in the United States. In Asian countries, ST59 is the most prevalent strain, especially in China, Taiwan, Vietnam and Singapore (12). Meanwhile, although cases of genetically identified USA300 CA-MRSA infection have been sporadically noted (13), the CA-MRSA strains reported in Japan are genetically rich in diversity.

Recently, it has been reported that the Japan-intrinsic CA-MRSA strain (CA-MRSA/J), which is a form of ST8 that differs from USA300, is prevalent (14). We herein report the clinical course of a patient with systematically disseminated...
CA-MRSA/J infection and describe the genotypic analysis.

**Case Report**

A 74-year-old woman with type 2 diabetes mellitus who worked at a stock farm was transferred to the emergency center of Tsuyama Central Hospital due to a one-week history of lumbar and extremity pain. She had never been abroad. She was in a semi-unconscious state (Glasgow Coma Scale, E4V4M5) upon arrival, and her vital signs were as follows: blood pressure, 101/65 mmHg; heart rate, 120 beats/min (regular); respiratory rate, 24 breaths/min; oxygen saturation, 97% (while receiving 100% fraction of oxygen at 10 L/min using a reservoir mask); body temperature, 36.1°C. A physical examination revealed macular erythroderma on the patient’s trunk and apparent inflammatory changes on her forearms, wrist, knee and ankle joints (Fig. 1). Laboratory testing showed a state of high inflammation (white blood cells, 16,800/mm³; C-reactive protein 37.6 mg/dL; procalcitonin, >10 ng/mL), renal dysfunction (creatinine, 2.04 mg/dL; blood urea nitrogen, 73.7 mg/dL), disseminated intravascular coagulopathy (DIC: platelet, 2.4×10⁷/mm³; fibrin degradation products, 30.4 μg/mL; D-dimer, 19.8 μg/mL; prothrombin time, 53%), elevated serum creatine kinase (3,294 IU/L) and lactic acidosis (pH, 7.28; lactate, 9.5 mmol/L; HCO₃⁻, 14.5 mmol/L). The Acute Physiology and Chronic Health Evaluation II and Sequential Organ Failure Assessment scores on admission were 30 and 12, respectively. The patient was admitted to the intensive care unit (ICU) with mechanical ventilator support.

After admission, the patient was in shock, and a high fever was observed. Her urine was cloudy, and Gram staining revealed many inflammatory cells phagocytizing Gram-positive cluster-forming cocci. Joint puncture was performed in the bilateral shoulder, elbow and knee joints. All specimens obtained from these joints were purulent, and the number of inflammatory cells was uncountable in the bilateral shoulders, 100,900/mm³ in the right elbow, 74,100/mm³ in the left elbow, 209,600/mm³ in the right knee and 233,600/mm³ in the left knee. Gram staining of the synovial fluid revealed Gram-positive cluster-forming cocci phagocytized by numerous inflammatory cells. Continuous drainage was performed in the bilateral knee joints, while the other infectious joints were treated with single aspiration. Whole body computed tomography (CT) did not show any other findings; however, magnetic resonance imaging (MRI)
revealed an abscess in the right greater psoas muscle and an epidural abscess at the lumbar level (Fig. 2). The aspiration samples obtained from these sites using CT-guided puncture were also purulent, and a drainage tube was placed in the right greater psoas muscle. An eye examination demonstrated petechial hemorrhage of the bilateral palpebral and bulbar conjunctiva, and a fundus examination by an ophthalmologist disclosed bilateral retinal hemorrhage with a soft exudate that was considered to be formed by emboli.

The skin manifestations on the patient’s bilateral forearms rapidly deteriorated. On arrival, both forearms and hands were reddish and swollen. On day 2, blisters formed and necrotic changes with internal hemorrhage were detected. The inflammatory changes finally peaked on day 9. Desquamation of both hands was observed on day 11 (Fig. 1).

Although infective endocarditis (IE) was clinically suspected, transthoracic echocardiograms performed on days 1, 2, 6 and 14 did not reveal any typical signs of IE. However, the patient satisfied one major criterion (a positive blood culture) and three minor criteria (a body temperature higher than 38.0°C; vascular phenomena: conjunctival hemorrhage and cerebral emboli; and immunologic phenomena: Roth spots on the fundus) of modified Duke’s criteria and was definitively diagnosed with IE. In addition, the patient was diagnosed with staphylococcal toxic shock syndrome (TSS) based on the CDC criteria modified in 1997: a high fever, shock state, diffuse rash, desquamation and multiple organ impairment.

Including all three sets of blood cultures, MRSA was detected in 13 different specimens (blood, nasal cavity, sputum, urine, bilateral synovial fluid of the shoulder, elbow and knee joints, lumbar vertebra and epidural abscesses and right major psoas pus). Under the diagnosis of systematically disseminated MRSA infection, combination therapy consisting of linezolid (600 mg every 12 hours, intravenously), clindamycin (600 mg every 8 hours, intravenously) and meropenem (0.5 g every 8 hours, intravenously) was initiated. Despite the administration of intensive care, the shock state persisted and multiple organ impairment, including disorders of the liver, kidneys and gastrointestinal and hematological systems, was observed. We administered polymyxin B immobilized column direct hemoperfusion with subsequent continuous renal replacement therapy to control the significant inflammatory state. The antibiotic...
and 18 weeks of antibiotic therapy was completed. The patient’s subsequent clinical course was relatively uneventful, cerebral emboli at the left corona radiata (Fig. 2), the level of creatine kinase gradually increased. Although treatment was again converted to linezolid on day 21, as the tent unconsciousness and respiratory failure. The antibiotic therapy was changed to daptomycin (350 mg every 24

therapy was changed to daptomycin (350 mg every 24 hours, intravenously) on day 10, and percutaneous dilatational tracheostomy was performed on day 12 due to persistent unconsciousness and respiratory failure. The antibiotic treatment was again converted to linezolid on day 21, as the level of creatine kinase gradually increased. Although follow-up MRI performed on day 23 revealed newly emerged cervical (C4-6) and lumbal (L2-4) spondylitis with cerebral emboli at the left corona radiata (Fig. 2), the patient’s subsequent clinical course was relatively uneventful, and 18 weeks of antibiotic therapy was completed. The patient survived; however, she became almost permanently bedridden and was transferred to a long-term care sanatorium.

Bacterial analysis

The antibiotic susceptibility pattern, which is characteristic to that of CA-MRSA, was the same among all strains: resistant to all β-lactams and gentamycin and sensitive to minocycline, clindamycin, levofloxacin, sulfamethoxazole/trimethoprim, arbekacin, vancomycin, teicoplanin and linezolid. Therefore, the involvement of a single CA-MRSA strain was suspected as the pathogen in this case.

The 11 isolates obtained from the patient’s blood, sputum, urine, synovial fluid (bilateral shoulders, elbows and knees), epidural abscess and right major psoas abscess were genetically analyzed. The isolates obtained from the nasal cavity and lumbar vertebra were not analyzed because the former was not considered to be directly pathogenic to the patient and the latter was obtained later in the clinical course. Mul-

Cattle-origin strain No.1

Cattle-origin strain No.2

tilocus sequence typing (MLST), polymerase chain reaction (PCR) of various pathogenic factors and pulsed-field gel electrophoresis (PFGE) were performed. A summary of the results of these tests is shown in Fig. 3. It was found that all of the MRSA isolates belonged to ST8, staphylococcal cassette chromosome mec (SCCmec) type IV and coagulase type (Coa) III and were negative for PVL (Panton-Valentine leukocidin) and ACME (arginine catabolic mobile element). The patterns of positive pathogenic factors were also the same: sec (staphylococcal enterotoxin C), selp (staphylococcal enterotoxin-like P), sell (staphylococcal enterotoxin-like L), sec (staphylococcal complement inhibitor), sak (staphylokinase precursor), tst-1 (toxic shock syndrome toxin-1), lukE/D (leukotoxin LukE/leukotoxin LukS) and ednA (gene for epidermal cell differentiation inhibitor toxin A). In addition, the PFGE pattern was identical in all strains. Based on these results, it was concluded that a single CA-MRSA clone caused the systemically disseminated infection.

We also conducted an epidemiological surveillance investigation at the patient’s farm six months after admission. Specimens were obtained from the mamma of 10 calves that were currently or had been suffering from mastitis. Only two specimens were positive for S. aureus; however, the genotype differed from that of the specimens isolated from the patient (Fig. 3).

Discussion

The final diagnosis in the present case was CA-MRSA bacteremia, pyogenic arthritis of the bilateral shoulder, elbow and knee joints, cervical and lumbor spondylitis, an epidural abscess at the lumbar level, a right major psoas abscess, severe soft tissue infection of the bilateral forearms, definitive IE, urinary tract infection, staphylococcal TSS, DIC and septic shock. It is possible that other joints, such as the wrists and ankles, were also involved, as these areas were anomalously swollen.

The primary infectious focus was unknown in this case. The patient worked at a stock farm, had frequent contact with cattle until admission and exhibited many small scars on her extremities on admission. Therefore, we presumed her extremities to be the primary site of infection.

The patient was critically ill; however, she was successfully saved, possibly due to the use of a fundamental approach, that is surgical drainage. As shown in this report, a single strain of CA-MRSA was detected in more than 10 body parts. We performed drainage in a total of eight sites of infection (the bilateral shoulder, elbow and knee joints, the epidural abscess and the right greater psoas abscess) in the early phase of the clinical course. In addition to providing proper antibiotic therapy, the use of surgical drainage at the appropriate time was essential for improving the prognosis.

According to a previous report that analyzed 21 clinical strains of CA-MRSA collected in Japan from 2003 to 2010, a major genotype of CA-MRSA/J is ST8/spa606(t1767)/agrI/SCCmecIV1/CoaIII, with divergence in the spa type (14). All investigated strains were found to be negative for PVL, ACME and S. aureus pathogenicity island (SaPI) carrying sek and seq and positive for sep (66.7%, 14/21) and SaPI carrying tst, sec and sel. Most strains were resistant to gentamicin (95.2%, 20/21), with a higher level of production of TSST-1 in CA-MRSA/J than in USA300. Based on the results of genotyping and MLST, an isolate obtained in the present case was proven to be genetically similar to ST8 CA-MRSA/J, not USA300 (Fig. 3).

It has been reported that livestock-associated MRSA is present in 88% of Dutch veal calf farms (15). Mastitis in cattle is an issue of critical importance to livestock breeders because all infected calves must be slaughtered. Cattle mastitis is generally caused by S. aureus, and we speculate the possibility that the present patient’s CA-MRSA infection was a result of a zoonotic infection.

The epidemiology and pathogenicity of the Japan-intrinsic CA-MRSA strain remain unclear. Further investigations of the clinical characteristics of this life-threatening pathogen and its allied species are required.

The authors state that they have no Conflict of Interest (COI).

References


© 2014 The Japanese Society of Internal Medicine
http://www.naika.or.jp/imonline/index.html