行政院國家科學委員會專題研究計畫成果報告

燙傷加護中心 CEFTAZIDIME 抗藥性綠膿桿菌感染之流行病學研究

Epidemiologic Study on Ceftazidime-resistant *Pseudomonas aeruginosa* Infections in an Infensive Care Burn Unit

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中文摘要

對多種藥物具抗藥性之綠膿桿菌長期存在於燒燙傷病人身上，在過去之文獻尚未看到，由本院近年在加護病房 3 **個燒燙傷病患身上可分離出** 39 **株此類細菌。我們以各種不同方法如血清型、抗藥型和基因型做這些細菌之分型發現有一菌株，屬於綠膿桿菌** 04 **造成群突發，由此菌株長期存活於此** 3 **病患之身上（血液、導管、傷口、痰等檢體），此** 3 **個病患在住院期間長期接受抗生素治療，呼吸器和各種導管，和過去幾年分離之菌株比較，我們發現此一引起群突發之菌株在** 1996 **年即在該加護病房內分離出。**

關鍵詞：綠膿桿菌、燒燙傷單位、菌株散播

ABSTRACT

Long-term colonization of various body sites with subsequent severe infections in burn patients by a multidrug-resistant *Pseudomonas aeruginosa* clone (resistant to piperacillin, cefoperazone, ceftazidime, aztreonam, imipenem, cefepime, cefpirome, ofloxacin, ciprofloxacin, minocycline, and aminoglycosides) has not been previously reported. Thirty-nine isolates of multidrug-resistant *P. aeruginosa* (resistant to ceftazidime and at least three of the agents listed above) recovered from various clinical samples of three patients in an intensive care burn unit, and seven preserved isolates recovered from six patients in other medical wards at National Taiwan University Hospital were studied for their epidemiological relatedness. The epidemic could be attributed to a multidrug-resistant *P. aeruginosa* clone belonging to serogroup O:F (serogroup O:4) by means of antimicrobial susceptibility, O-serogrouping, and random amplified polymorphic DNA (RAPD) patterns generated by arbitrarily primed PCR (APPCR) of the isolates. The epidemic strain persisted in the three patients for weeks to months; in the meanwhile these patients had received multiple antimicrobial agents for management of intervenient episodes of invasive infections (bacteremia, ventilator-associated pneumonia, and/or catheter-related sepsis) caused by this strain, as well as concomitant infections due to other organisms. The strain had been isolated only once previously from a burn patient of the unit in December 1996. The present study describing a small outbreak due to *P. aeruginosa* documents that a single clone of multidrug-resistant *P. aeruginosa* can cause long-term persistence in different body sites of burn patients and subsequently result in various severe infections.

Keywords: *Pseudomonas aeruginosa*, burn unit, clonal dissemination

INTRODUCTION

Despite advances in surgical care and the introduction of a wide variety of antimicrobial agents with antipseudomonal activity, life-threatening infection caused by *Pseudomonas aeruginosa* continues to be a common complication in burn patients and

contributes substantially to burn-related morbidity and mortality worldwide. Multidrug-resistant *P. aeruginosa* have been frequently reported as the cause of nosocomial outbreaks of infection in burn units or as colonizers of the wounds of burn patients. Also, the long-term colonization by more than one *P. aeruginosa* clone in the respiratory tracts of patients with cystic fibrosis or bronchiectasis has been well demonstrated by various genotypic and phenotypic methods. However, the situation of a multidrug-resistant *P. aeruginosa* clone colonizing various body sites of burn patients for weeks and months and causing intervenient episodes of severe infection has not been previously described.

MATERIALS AND METHODS

Background. The intensive care burn unit of National Taiwan University Hospital has 8 single-bed intensive care rooms. Three unusual isolates of *P. aeruginosa* recovered from various clinical specimens of two patients (patients 2 and 4), were resistant to 12 antimicrobial agents (cefoperazone, ceftazidime, aztreonam, piperacillin, ticarcillin/clavulanic acid, imipenem, minocycline, gentamicin, tobramycin, amikacin, ofloxacin, and ciprofloxacin) using the routine disk diffusion method. It was noted upon reviewing microbiological records that, only one *P. aeruginosa* strain with the same antibiotype had been previously isolated, that from a patient who had been hospitalized from the intensive care burn unit in December 1996.

Epidemiological surveillance. After the infections with the multidrug-resistant *P. aeruginosa* strains were discovered, bacterial cultures for multidrug-resistant *P. aeruginosa* isolates (resistant to ceftazidime and at least three of the agents listed above) were performed in various samples from all patients in the burn unit. Environmental culture surveillance was undertaken to detect the presence of multidrug-resistant *P. aeruginosa* from various sources: 100 ml of water from sink faucets and swabs of sink surfaces in each patient's room. Swab specimens from the hands and nasal nares of the physicians and nurses in the unit were also collected for cultures due to the concomitant occurrence of an outbreak due to oxacillin-resistant *Staphylococcus. aureus*.

Determination of O-serogroup. Group antisera against 14 O-serogroup antigens, designated as A to N, were purchased from Denka Seiken Co., Ltd. (Tokyo, Japan).

Random amplified polymorphic DNA (RAPD) patterns. RAPD patterns generated by arbitrarily primed PCR (APPCR) was performed essentially as described before. Two arbitrary oligonucleotide primers were used: M13: 5'-TTATGTAAAACGACGGCCAGT-3' (Gibco BRL Products, Gaithersburg, Md.) and H5: 5'-AGTCGTCCCC-3' (OPERON Technologies, Inc., Alameda, Calif.). RAPD patterns of the isolates with a difference of one or more discrete bands were considered different, otherwise were considered identical.

RESULTS

Characteristics of patients. During the two months (April and May) of investigation, 16 patients were treated in the intensive care burn unit. A total of 47 isolates of *P. aeruginosa* were included in this study: 40 from various clinical samples from four patients in the burn unit and seven from six patients in other medical wards. The four burn patients all had more than a 60% total-body-surface-area (TBSA) burn wound and had been exposed to various invasive procedures (central venous catheter insertion

and ventilator use) during their stay in the burn unit. None of these patients received hydrotherapy. These patients all had been treated with various β-lactams, including those with antipseudomonal activity (piperacillin, ceftazidime, and imipenem) and aminoglycosides before the acquisition of *P. aeruginosa* infection or colonization. In addition to the colonization or infection of the burn wounds of the four patients by the epidemic multidrug-resistant *P. aeruginosa* strain, other complicated infections caused by this organism did occur: patient 2 had bacteremia and ventilator-associated pneumonia, and patients 3 and 4 both had central venous catheter-related sepsis and ventilator-associated pneumonia. After the notification of the presence of the multidrug-resistant *P. aeruginosa*, these four burn patients received a wide array antimicrobial agents, including third-generation cephalosporins, aminoglycosides, and ciprofloxacin, which were prescribed for the treatment of concomitant infections caused by pathogens other than *P. aeruginosa*. Patient 2 died of respiratory failure due to a fulminant pneumonia caused by oxacillin-resistant *S. aureus*. Patients 3 and 4 both survived, although strains of *P. aeruginosa* were repeatedly isolated from different clinical specimens during the course of intensive burn unit care.

Susceptibility testing. All isolates from patients 2 (except for isolate B1), 3 (isolates C1 to C15), and 4 (except for isolates D2, D13, D16, and D19) had MIC values to all antimicrobial agents tested identical with those of isolate A, except aztreonam with which no more than one twofold dilution discrepancy of MIC values (MICs, 32 to 64 μg/ml) was found among these isolates. A total of 12 antibiotypes were found (Table II): six antibiotypes were found among the 40 isolates recovered in the burn units and the other six were discovered among the seven isolates from patients on medical wards (Table I).

O-serogrouping. The epidemic multidrug-resistant strains all belonged to serogroup O:F (serogroup O:4). Isolates belonging to the same antibiotype had identical serogroups. However, isolates D2, D16, and J, which had the same serogroup (serogroup O:E), had different antibiotypes. Isolates G1 and G2 from patient 7 had the same antibiotype and both belonged to the O-serogroup A.

RAPD patterns. The epidemic multidrug-resistant serogroup O:F *P. aeruginosa* isolates all possessed an identical RAPD pattern. Isolates with the same antibiotype and O-serogroup also had the same RAPD pattern.

Infection control measures. Sampling cultures from healthcare workers were positive for oxacillin-resistant *S. aureus* in three physicians and two nurses but all negative for *P. aeruginosa.* None of the environmental sites were positive for *P. aeruginosa*. Environmental decontamination measures were not undertaken. The physicians in the unit were not informed of restriction of use of third-generation cephalosporins, aztreonam, or imipenem or changing dosages of antimicrobial agents administered.

DISCUSSION

The present study, using phenotypic and genotypic characterization of 40 isolates of multidrug-resistant *P. aeruginosa* recovered from four patients in an intensive care burn unit, disclosed two important points. First, more than one clone of multidrug-resistant *P. aeruginosa* can colonize not only burn wounds but also other body sites of burn patients. Second, a

single clone of multidrug-resistant *P. aeruginosa* can persist in different body sites of burn patients for weeks and months and subsequently cause various severe infections.

The transfer route of the O:F strain was not identified. Previous studies suggested that culture surveys to identify reservoirs of *P. aeruginosa* in burn units have yielded positive cultures from sinks and hydrotherapy equipment. However, only hydrotherapy equipment is strongly linked to the epidemic of *P. aeruginosa* burn wound infections. Consequently, dispersal of *P. aeruginosa* from colonized or infected patients in the burn unit will result in further contamination of the environment of the unit as well as the hands of medical personnel. In the present investigation, all cultures of the environmental samples and hands and nasal swabs of medical personnel were negative for the epidemic strain. Colonization of the rectum by *P. aeruginosa* in burn patients is considered to be a potential source of subsequent burn wound infection. Only two of the cultures of stool samples obtained from the study patients yielded the epidemic strain during surveillance of the outbreak. Whether the epidemic *P. aeruginosa* strain of the two patients was acquired from the alimentary tract or were merely contaminants in the stool specimens from nearby infected skin is unclear, because cultures of stool or rectal swab specimens before the first positive epidemic strain from other body sites were not taken

The present study, using phenotypic and molecular studies, highlights that a single clone of pan-drug-resistant *P. aeruginosa* can persist in different body sites of burn patients for weeks and months and subsequently cause various severe infections and the difficulty in duly eradicating the pan-drug-resistant strains from these patients. The extensive use of third-generation cephalosporins in the unit is probably responsible for the emergence and selection of this pan-drug-resistant strain.

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