How important is patient-to-patient transmission in extended-spectrum β-lactamase *Escherichia coli* acquisition

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Background: Extended-spectrum β-lactamase (ESBL)-producing *Escherichia coli* is an emerging pathogen. The causal role of antibiotic selective pressure versus patient-to-patient transmission has not been assessed. The objective of this study was to quantify the amount of patient-to-patient transmission among patients who acquire an ESBL-producing *E coli* infection using perianal surveillance cultures in an intensive care unit (ICU) population.
Methods: A prospective cohort of patients admitted between September 1, 2001, and September 1, 2004, to the medical and surgical ICUs at a tertiary care hospital was studied. Patients had perianal cultures on admission, weekly, and upon discharge. Strain typing by pulsed-field gel electrophoresis (PFGE) and epidemiologic criteria were used to quantify the amount of patient-to-patient transmission.
Results: There were 1806 patients admitted to the ICUs. There were 74 patients who had ESBL-producing *E coli* on admission to the ICU and 23 patients who acquired ESBL-producing *E coli*. Among these 23 patients, there were 14 PFGE types, and 3 (13%) patient acquisitions were defined as patient-to-patient transmission by similar PFGE type and overlapping time in the hospital.
Conclusion: Our data suggest that patient-to-patient transmission is not an important cause of the acquisition of ESBL-producing *E coli* colonization in the ICU setting. (Am J Infect Control 2007;35:97-101.)

Antibiotic-resistant gram-negative bacteria are an emerging problem that infection control practitioners, hospital epidemiologists, clinicians, and hospital administrators are struggling to control.[1,2] *Escherichia coli* producing extended-spectrum β-lactamases (ESBL) are an important emerging gram-negative bacteria because of lack of consistent detection in the clinical laboratory,[3,4] limitations of effective antibiotic therapy,[1,5] and adverse patient outcomes.[6,7]

Control of antibiotic-resistant gram-negative bacteria, including ESBL-producing bacteria, requires the understanding of the relative causal importance of (1) the organism-specific proportion of antibiotic resistance attributable to antibiotic use (ie, attributable fraction) and (2) the organism-specific attributable fraction because of patient-to-patient transmission. At present, studies attempting to measure the attributable fraction because of patient-to-patient transmission of antibiotic-resistant gram-negative bacteria have been limited. In addition, they have been of small sample size, have often only been performed during outbreaks thus limiting their generalizability, and have focused on clinical cultures positive for ESBL-producing bacteria. No studies have focused on gastrointestinal tract colonization acquisition of ESBL-producing *E coli*.

The objective of this study was to quantify the amount of patient-to-patient transmission among patients who acquire ESBL-producing *E coli* using perianal surveillance cultures in an intensive care unit (ICU) population.

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Supported by the National Institutes of Health grants K23 AI01752-01A1 and R01 AI60859-01A1.

0196-6553/$32.00

doi:10.1016/j.ajic.2006.09.011

METHODS

Study design and patient population

This study was approved by the Institutional Review Board of the University of Maryland, Baltimore. This study utilized a prospective cohort of adult patients admitted to the medical ICU (MICU) and surgical ICU (SICU) of the University of Maryland Medical Center (UMMC) between September 1, 2001, and September 1, 2004. The UMMC is a tertiary care facility in Baltimore, Maryland. The MICU is a 10-bed, private room unit providing care to patients who have acute or potentially life-threatening medical conditions including hematologic and other malignancies. The SICU is a

19-bed, private room unit providing care to adult patients with solid organ transplantation and abdominal, genitourinary, orthopedic, and otolaryngologic surgery.

During the study period, patients in the MICU and SICU had admission, weekly (every Wednesday), and discharge perianal cultures performed. The culturing technique involved swabbing the perianal area in a circular motion. Samples for cultures were obtained by nurses and nursing assistants (extenders) as part of an ongoing vancomycin-resistant Enterococci (VRE) active surveillance program. At the same time, screening for ESBL-producing organisms of perianal cultures was performed. During the study period, infection control monitoring and control charting for clinical cultures positive for ESBL-producing bacteria among MICU and SICU patients revealed no evidence of an outbreak.

We defined admission-positive patients as patients who had admission cultures positive for ESBL-producing *E coli* bacteria. We defined acquisition-positive patients as patients who had admission cultures negative for ESBL-producing *E coli* and had either a subsequent weekly or discharge culture positive for ESBL-producing *E coli*.

Microbiologic methods

For isolation of ESBL-producing *E coli*, swabs were plated onto MacConkey agar (Remel, Lenexa, KS) with 1 μg/mL of ceftazidime added to the cooled agar before plates were poured. Plates were incubated at 37°C for 24 to 48 hours. Lactose fermenting colonies growing on the ceftazidime containing plates that were identified as *E coli* with API 20E Identification Strips (BioMerieux Vitek Inc., Hazelwood, MO) underwent ESBL confirmatory testing by disk diffusion in accordance with Clinical Laboratory Standards Institute (CLSI) guidelines.[8]

Molecular methods and definition of patient-to-patient transmission

Pulsed-field gel electrophoresis (PFGE) was performed as previously described with the exception of run parameters.[9] Briefly, genomic DNA was digested with *Xba*I according to the manufacturer's recommendations (New England Biolabs, Beverly, MA) Genomic DNA was separated in 1% agarose using a CHEF-DR II (Bio-Rad, Richmond, CA). Electrophoresis was performed at 200 V for 22 hours, with an initial switch time of 5 seconds and the final switch time of 40 seconds. Gels were stained with ethidium bromide and photographed using the Gel Doc (Bio-Rad, Hercules, CA). Interpretation was performed using Fingerprinting II software (Bio-Rad). A dendrogram comparing all 97 isolates was constructed using Dice coefficient and unweighted pair group method with arithmetic mean with a position tolerance of 1. Isolates with >90% similarity were considered similar according to Tenover criteria.[10]

We defined patients who acquired ESBL-producing *E coli* as having acquired because of patient-to-patient transmission if (1) their isolate was defined as similar based on the PFGE type and (2) they were defined as epidemiologically related based on any overlap in hospital length of stay, ie, a patient was defined as to have acquired because of patient-to-patient transmission from another patient if their PFGE type was similar and their respective hospital lengths of stay overlapped the same portion of the calendar.

RESULTS

There were 1806 patients admitted to the MICU and SICU during the 3-year study period who had admission and discharge cultures obtained. Compliance with obtaining perianal surveillance cultures (admission and discharge) was greater than 90% on average. Mean demographics of these patients are as follows: age, 55.7 years; female, 46%; Charlson score, 2.3; chronic disease score, 7.8; average time in hospital before ICU admission, 2.6 days; average numbers of days in the ICU, 6.8 days.

Ninety-seven patients had an ESBL-producing *E* coli, of which 74 had an ESBL-producing *E coli* on admission to the ICU and thus were defined as admission-positive patients. There were 23 patients who had an admission culture negative for ESBL-producing *E coli* and a subsequent weekly or discharge culture positive for ESBL-producing *E coli* and thus were defined as patients who acquired ESBL-producing *E coli*. Of the 23 patients who acquire ESBL-producing *E coli*, only 2 patients were known to be ESBL colonized based on previous positive clinical cultures. During the same time period, 27 patients acquired ESBL-producing *Klebsiella pneumoniae*.

Molecular typing discriminated the 97 ESBL-producing *E coli* into 41 unique PFGE types, which is shown in Fig 1. Among the 23 acquisition patients, there were 14 unique PFGE types, and 3 isolates had identical PFGE type and overlapping length of stay in the hospital (Fig 2). Thus, based on our definitions, 3 (13%) of 23 acquisitions were believed to have acquired their ESBL-producing bacteria by patient-to-patient transmission.

Two major PFGE groups (PFGE types 6 and 27) were seen. However, type 6 only had 1 acquired isolate that had overlap in hospital stay and was defined as patient-to-patient transmission. For PFGE group 27, all cultures were admission cultures.

DISCUSSION

In this study, we quantified the amount of patient-to-patient transmission of ESBL-producing *E coli* that

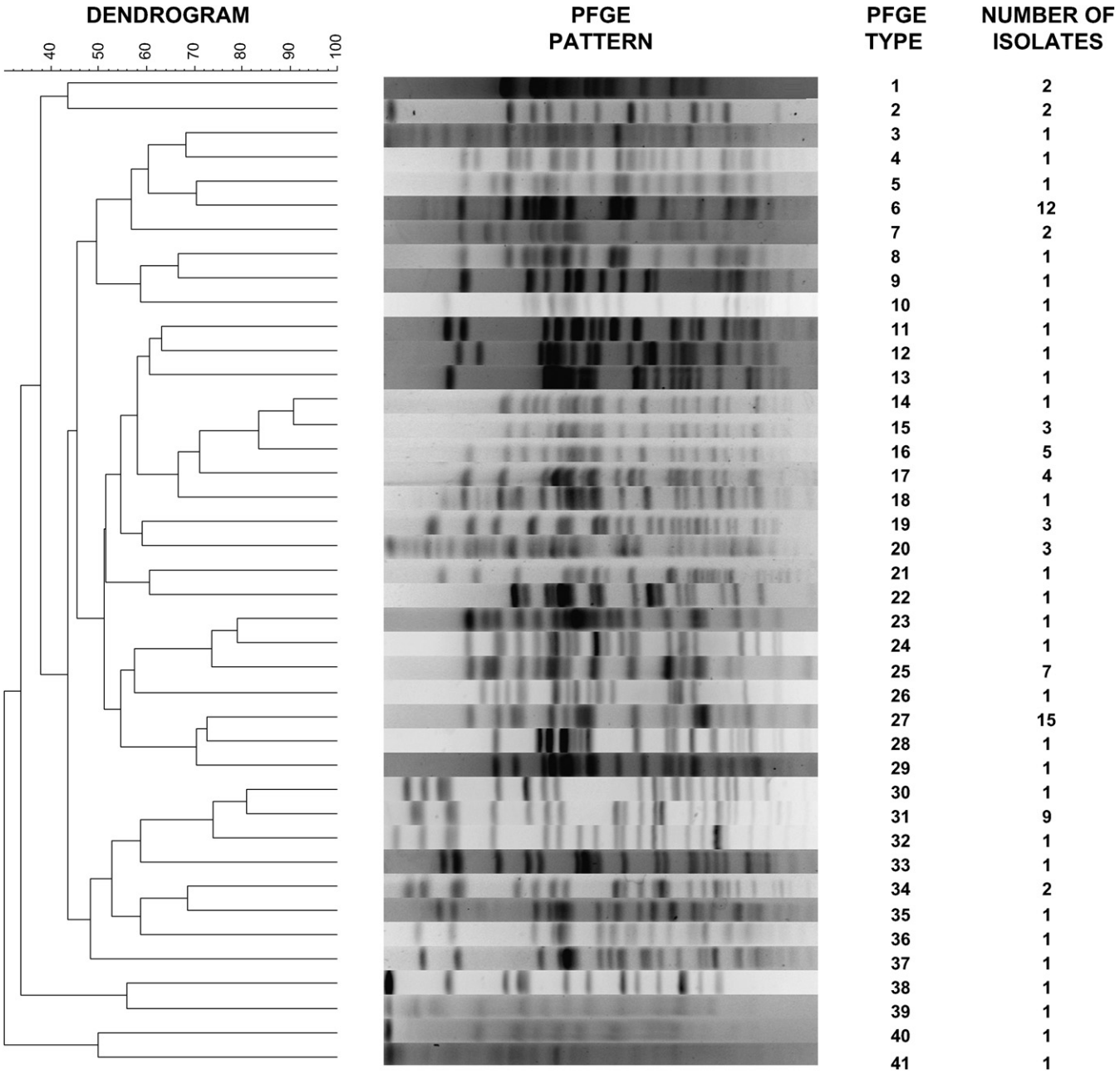

Fig 1. PFGE dendrogram of 41 PFGE types of ESBL-producing *E coli* with the number of isolates within each type. Each row contains a representative isolate from that PFGE type. Greater than 90% was considered similar PFGE type.

occurred in a tertiary care hospital over a 3-year period. To our knowledge, we are the first to focus this quantification for ESBL-producing *E coli* in the setting of gastrointestinal colonization. We found that, of the 23 patients who acquired colonization with ESBL-producing bacteria, 3 (13%) had patient-to-patient transmission, defined as similar PFGE type and epidemiologic hospital time overlap.

Although there have been previous studies looking at patient-to-patient transmission of ESBL-producing bacteria, none of those have studied patient-to-patient transmission of ESBL-producing *E coli* colonization. Gardam et al performed a cohort study on colonization and clinical infection among organ transplantation patients, and their results suggested little patient-to-patient transmission of third-generation cephalosporin-resistant *Enterobacteriaceae*.[11] In their study, among the 69 patients who had a resistant *Enterobacteriaceae* (both clinical culture and active surveillance), they found 66 unique PFGE types, and only 2 of their 69

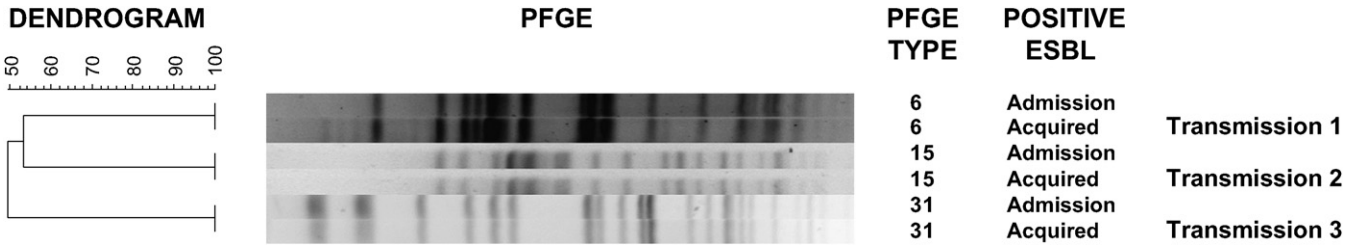

Fig 2. PFGE dendrogram of ESBL-producing *E coli* of 3 patient-to-patient transmissions defined by similar PFGE patterns and hospital overlap. The acquired isolate is paired with the admission isolate of the same PFGE type.

patient isolates were ESBL-producing *E coli*. Decre et al[12] analyzed risk factors for ESBL-producing *Klebsiella* and found a large amount of patient-to-patient transmission. Their study, in contrast to ours, was performed in an outbreak setting and focused on the acquisition of *Klebsiella*, a genetically different member of the *Enterobacteriaceae* family.[12]

We believe from our study that the amount of patient-to-patient transmission of ESBL-producing *E coli* in an ICU setting is low. However, our compliance rate with obtaining surveillance cultures was 90%, and, thus, we may have underestimated the amount of patient-to-patient transmission. In addition, there is no universal definition of ''epidemiologically related.'' We chose a definition that involved the need for hospital length of stay to overlap. However, there may have been some acquisition patients who had similar PFGE types to other patients who did not have temporal hospital length of stay overlap but did acquire the organism because of patient-to-patient transmission through longer term colonization of health care workers or environmental products or services. Of our 23 acquisition patients, there were 9 patients who had similar PFGE types. This suggests that, if patient-to-patient transmission was defined based on only the PFGE types, likely an overestimate, 9 (39%) of the acquisitions may have been due to patient-to-patient transmission. This percentage may highlight and establish an upper level confidence interval of the amount of patient-to-patient transmission.

Although we did detect 3 patient-to-patient transmissions, we might be underestimating patient-to-patient transmission because of the transmission of plasmids. Thus, organisms that are unrelated by PFGE analysis may have had plasmid transfer of ESBL-resistance mechanisms. If plasmid transfer of resistance mechanisms occurs, this would also contribute to an underestimate of the amount of patient-to-patient transmission. Future work on plasmid resistance mechanism transfer needs to be performed. Another limitation of the study is that the sensitivity of perianal culture compared with stool culture for ESBL-producing bacteria is unknown. However, the sensitivity of perianal culture compared with stool culture for fluoroquinolone-resistant *E coli* is 90%.[13]

We caution that our results should likely not be generalized to other ESBL-producing bacteria that may have different attributable fractions because of patient-to-patient transmission, and, thus, other members of the Enterobacteriaceae family should be studied. In particular, *Klebsiella pneumoniae* and *Klebsiella oxytoca* need to be studied because they are a major contributor to ESBL-producing bacteria.[1,12,14] In addition, the amount of transmission of antibiotic-resistant bacteria depends on the colonization pressure.[15,16] Thus, our results may or may not be generalizable to institutions with a higher prevalence of ESBL-producing *E coli*.

In conclusion, our study suggests that patient-to-patient transmission is not an important cause of the acquisition of ESBL-producing *E coli* colonization acquisition in the ICU in a tertiary care hospital in a nonoutbreak setting. Future studies examining the risk factors for ESBL-producing *E coli* colonization and acquisition need to be done. Risk factors that should likely be explored include antibiotic use and comorbid conditions. These future studies could lead to interventions aimed at antibiotic usage that will curb the emergence of ESBL-producing *E coli*.

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