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Clinical and Microbiologic Characteristics of Cephalosporin-Resistant *Escherichia coli* at Three Centers in the United States

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We investigated the clinical and microbiologic features of 300 cases of cephalosporin-resistant *Escherichia coli* producing extended-spectrum β-lactamase (ESBL) or plasmid-mediated AmpC β-lactamase (pAmpC) at three medical centers in the United States. Solid-organ malignancy, connective tissue disease, and a recent history of surgery were more common among pAmpC-producing cases ($n = 49$), whereas urinary catheter at enrollment, diabetes, and hospitalization in the past year were more common among ESBL-producing cases ($n = 233$). The factors independently associated with clinical outcome were the following: the presence of cardiovascular disease (odds ratio [OR], 2.88; 95% confidence interval [CI], 1.29 to 6.43), intra-abdominal infection (OR, 6.35; 95% CI, 1.51 to 26.7), other or multiples sources of infection (OR, 8.12; 95% CI, 2.3 to 28.6), age of 65 years or greater (OR, 0.43; 95% CI, 0.2 to 0.95), favorable baseline health status (OR, 0.39; 95% CI, 0.16 to 0.95), and appropriate empirical antimicrobial therapy given in the first 72 h (OR, 0.42; 95% CI, 0.20 to 0.88). β-Lactamase genes responsible for cephalosporin resistance were identified in 291 cases. CTX-M-type ESBLs accounted for 72.0%. Of those, 88.0% were CTX-M-15. The next most common type was CMY-type pAmpC (16.7%), followed by SHV- and TEM-type ESBLs (6.3 and 1.3%, respectively). Seven cases (2.3%) had KPC-type β-lactamase. Ertapenem, imipenem, meropenem, doripenem, piperacillin-tazobactam, amikacin, nitrofurantoin, and tigecycline were highly active, with greater than 90% of the isolates being susceptible. Cefepime was less active, with only 74.2% being susceptible due to the predominance of CTX-M-15. These findings have implications in the selection of appropriate empirical therapy when infection due to cephalosporin-resistant *E. coli* is suspected.

Cephalosporin-resistant *Escherichia coli* producing extended-spectrum β-lactamase (ESBL) or plasmid-mediated AmpC β-lactamase (pAmpC) commonly occurs in health care settings as well as in the community (7, 11). The rising incidence of these organisms, coupled with their spread into nursing homes and the community, has implications for the empirical management of patients presenting with infections in which *E. coli* is suspected as the causative pathogen.

CTX-M-type ESBLs are replacing the conventional TEM- and SHV-type ESBLs in *E. coli* worldwide (11, 13). While the United States initially appeared to be spared from this epidemic (1), it has subsequently been shown in surveillance and single-center studies that CTX-M-type ESBLs are becoming common in U.S. hospitals as well (8, 9, 12, 17). In Pittsburgh, more than 70% of ESBL-producing *E. coli* strains isolated between 2007 and 2008 were found to produce CTX-M-type ESBLs (15).

Infections due to pAmpC-producing *E. coli* are likely common but underrecognized due to the lack of standardized detection methods (16). The most commonly observed pAmpC in *E. coli* is the CMY type (7). In addition to penicillins, cephalosporins, and aztreonam, pAmpC-producing *E. coli* is often resistant to β-lactam–β-lactamase inhibitor combinations as well.

Given the paucity of clinical and microbiologic data regarding cephalosporin-resistant *E. coli* from the United States, we conducted this multicenter study to determine the risk factors for acquisition and predictors of clinical outcome associated with these organisms. In addition, we elucidated the epidemiology of the relevant β-lactamases and their correlation with antimicrobial susceptibility.

MATERIALS AND METHODS

Patients. Three medical centers participated in this study: the University of Pittsburgh Medical Center (Pittsburgh, PA), Henry Ford Hospital (Detroit, MI), and the University of Texas Health Science Center at San Antonio (San Antonio, TX). Ethical approval was obtained from the institutional review board at all three sites. Patients who had a clinical culture positive for cephalosporin-resistant *E. coli* between September 2008 and June 2010 were prospectively identified and included in this observational, cohort study. Criteria for inclusion were the following: (i) *E. coli* was identified as producing ESBL or otherwise was nonsusceptible to ceftriaxone, and (ii) medical records were available for review for at least 3 days from the date the culture was taken. Since a substantial portion of the cases for which urine was the source of *E. coli* represented colonization rather than infection and would not contribute to the clinical outcome analysis, nonurinary cases were included first, followed by urinary cases from the study period. A patient could be included in the study only once.

Clinical data collection. Clinical information collected included demographics, type of infection, underlying medical conditions, previous contact with the health care system, previous antimicrobial use, the presence of indwelling catheters, the duration of hospital stays before enrollment, pertinent culture results, antimicrobial treatment given, temperature, white blood cell count for up to 5 days, and survival at 28 days after enrollment. Each case was classified as hospital acquired, health care as-

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sociated, or community acquired in accordance with the definition of Friedman et al. (5). The clinical outcome of the cases was analyzed according to the antimicrobial therapy given, the source of infection, the site of acquisition, and the mechanism of cephalosporin resistance. Antimicrobial therapy was classified into initial empirical and definitive therapy, with the former defined as the initial therapy provided within 72 h after the recovery of *E. coli* and the latter defined as therapy provided after the results of susceptibility tests had been reported. Therapy was considered appropriate when the isolate was reported as susceptible to the agent by the clinical microbiology laboratory. Isolates confirmed as producing ESBL were considered resistant to penicillins, cephalosporins, and aztreonam regardless of the actual susceptibility, since all of the participating sites used the conventional breakpoints and recommendations by the Clinical and Laboratory Standards Institute (CLSI; prior to 2010) throughout the study period (2). The source of infection was determined as respiratory tract infection (including pneumonia), urinary tract infection, surgical site infection, intra-abdominal infection, line-related infection, or primary bacteremia using the definitions of the Centers for Disease Control and Prevention (CDC) (6). The overall baseline health status at the time of the positive culture was assessed using a modified McCabe-Jackson scoring system: A, likely survival for <4 days; B, ≥4 days but <1 month; C, ≥1 month but <5 years; and D, ≥5 years (10). Clinical response was defined by meeting all of the following criteria for a continuous 24-h period before or at 96 h from the time the first culture with *E. coli* was taken: (i) temperature, 36.0 to 37.9°C; (ii) white blood cell count, 4,000 to 10,900/μl; (iii) no requirement for vasopressors; (iv) systolic blood pressure, >90 mmHg; and (v) absence of relevant clinical symptoms and/or signs. A case was documented as clinical failure when any of these criteria was not met. The primary endpoints for the outcome analysis were clinical response at 96 h and survival at 28 days.

Microbiology procedures. All *E. coli* isolates were sent to the research laboratory at the University of Pittsburgh for further analysis. The MICs of β-lactam and non-β-lactam antimicrobials were determined using commercially available dry plates (Sensititre GN4F; TREK Diagnostic Systems, Cleveland, OH). Susceptibilities of the study isolates to cefazolin, ceftriaxone, ceftazidime, aztreonam, ertapenem, imipenem, and meropenem were compared using the CLSI 2009 and 2010 breakpoints (2, 3). PCR analysis for the detection of TEM-, SHV-, CTX-M-, CMY-, FOX-, and KPC-type β-lactamase genes were conducted as described elsewhere (4, 15). All PCR products were sequenced for SHV-, CTX-M-, CMY-, and KPC-type β-lactamase genes. TEM-type β-lactamase genes were sequenced only when the other PCR and sequencing results did not reveal a mechanism for cephalosporin resistance, since *E. coli* commonly possesses the TEM-1 gene, which encodes a non-ESBL.

Statistical analysis. Univariate analyses were performed separately for each of the variables. Odds ratios (ORs) and 95% confidence intervals (CIs) were calculated for binomial variables. *P* values were calculated by the use of the chi-square test and median unbiased estimate for categorical variables. All variables with *P* values of less than 0.2 in the univariate model were eligible for inclusion in the multivariable model. The analysis was a logistic regression model that was performed in a stepwise manner with a stay criteria of $P < 0.05$. Upon the determination of the initial model, variables that were not significant were removed from eligibility and the stepwise model was run again to determine a final model. All tests were two-tailed. SAS version 9.2 was used for the analysis (SAS Institute, Cary, NC).

RESULTS

Demographics and risk factors. A total of 300 cases were included in the study. The demographics and clinical characteristics of 49 cases with pAmpC-producing *E. coli* and 233 cases with ESBL-producing *E. coli* are presented in Table 1. The former group included 28 cases of infection and 21 cases of colonization, whereas the latter group included 152 cases of infection, 75 cases of colonization, and 6 indeterminate cases. Cases with isolates producing KPC ($n = 7$) and both pAmpC and ESBL ($n = 2$) were excluded from this analysis. The majority of patients were female in both groups. Approximately a third of the cases in both groups represented colonization only, mostly in the urine. The majority of the cases were health care associated, whereas approximately a third and less than 10% were hospital acquired and community associated, respectively (Table 1).

In the multivariate analysis, solid-organ malignancy (OR, 3.32; 95% CI, 1.42 to 7.75; $P = 0.06$), connective tissue disease (OR, 8.21; 95% CI, 1.49 to 45.4; $P = 0.016$), and surgery in the past 90 days (OR, 2.33; 95% CI, 1.12 to 4.83; $P = 0.024$) were independent risk factors for pAmpC cases but not for ESBL cases. On the other hand, independent risk factors for ESBL cases that were not significant for pAmpC cases included the presence of a urinary catheter at enrollment (OR, 0.18; 95% CI, 0.06 to 0.51; $P = 0.001$), diabetes (OR, 0.44; 95% CI, 0.20 to 0.96; $P = 0.039$), and hospitalization in the past year (OR, 0.33; 95% CI, 0.15 to 0.72; $P = 0.006$).

The 7 KPC cases then were compared to the ESBL cases. Due to the small number of cases, only univariate analysis was conducted. In this analysis, a history of organ transplantation (OR, 7.54; 95% CI, 1.58 to 35.96; $P = 0.01$), immunosuppressive therapy in the prior 30 days other than corticosteroids (OR, 10.2; 95% CI, 2.09 to 49.83; $P = 0.004$), and liver disease (OR, 8.41; 95% CI, 1.75 to 40.37; $P = 0.008$) were significant risk factors for KPC cases.

Predictors of clinical outcome. Predictors of clinical outcome at 96 h and mortality at 28 days were analyzed. Clinical outcome at 96 h could be assessed for 181 cases that represented infection (Table 2). The variables independently associated with clinical failure were cardiovascular disease (OR, 2.88; 95% CI, 1.29 to 6.43; $P = 0.01$), intra-abdominal infection (OR, 6.35; 95% CI, 1.51 to 26.7), and other or multiples sources (OR, 8.12; 95% CI, 2.30 to 28.6) of infection ($P = 0.008$). Factors that were protective included an age of 65 years or older (OR, 0.43; 95% CI, 0.20 to 0.95; $P = 0.04$), favorable overall baseline health status as defined by a modified McCabe-Jackson score of D (OR, 0.39; 95% CI, 0.16 to 0.95; $P = 0.04$), and appropriate antimicrobial given in the first 72 h (OR, 0.42; 95% CI, 0.20 to 0.88; $P = 0.02$).

Mortality at 28 days was 11.1% among 144 cases for which the information was available. In the univariate analysis, dialysis in the past 90 days (OR, 6.65; 95% CI, 1.85 to 23.84; $P = 0.004$), attendance at a dialysis clinic in the 30 days prior to enrollment (OR, 5.49; 95% CI, 1.18 to 25.67; $P = 0.03$), the presence of a vascular catheter at enrollment (OR, 5.21; 95% CI, 1.69 to 16.02; $P = 0.004$), hospital-acquired infection (OR, 5.21; 95% CI, 1.69 to 16.02; $P = 0.004$), liver disease (OR, 4.52; 95% CI, 1.44 to 14.23; $P = 0.01$), intra-abdominal infection (OR, 10.1; 95% CI, 1.23 to 83.5), and other or multiple sources (OR, 19.1; CI, 3.57 to 102) of infection ($P = 0.04$) were significantly associated with mortality at day 28. The predictors for survival at 28 days were carbapenem use for at least 1 day any time after infection (OR, 0.2; 95% CI, 0.07 to 0.64; $P = 0.006$) and a modified McCabe-Jackson score of D (OR, 0.1; 95% CI, 0 to 0.59; $P = 0.007$). Multivariate analysis was deferred due to the small number of patients who died.

Mechanism of cephalosporin resistance. The β-lactamases responsible for cephalosporin resistance could be identified by PCR and sequencing experiments in 291 cases. CTX-M-type ESBLs accounted for 72.0% of them (Table 3). One hundred ninety-one isolates (63.7%) carried CTX-M-1-group genes, accounting for 88.0% of the CTX-M-type ESBLs. All of the CTX-M-1-

TABLE 1 Demographics and clinical characteristics of patients with infection or colonization due to pAmpC β-lactamase-producing or ESBL-producing *E. coli*

Parameter	Result by analysis type					
	Univariate				Multivariate	
	No. (%) of pAmpC producers (n = 49)	No. (%) of ESBL producers (n = 233)	Unadjusted OR (95% CI)	P value	Adjusted OR (95% CI)	P value
Demographics						
Male gender	18 (36.7)	93 (40.4)	0.86 (0.45, 1.62)	0.63		
Age ≥65 yr	20 (40.8)	126 (54.3)	0.58 (0.31, 1.08)	0.09		
Race				0.13		
Black	7 (15.2)	66 (30.1)	Baseline			
White	32 (69.6)	124 (56.6)	2.43 (1.02, 5.81)			
Other	7 (15.2)	29 (13.2)	2.28 (0.73, 7.08)			
Location of acquisition				0.84		
Community associated	4 (8.3)	22 (9.4)	Baseline			
Health care associated	26 (54.2)	135 (57.5)	1.06 (0.34, 3.33)			
Hospital acquired	18 (37.5)	78 (33.2)	1.27 (0.39, 4.14)			
Health care-associated risk factor						
Any hospitalization in past year	32 (65.3)	173 (80.1)	0.47 (0.24, 0.92)	0.03	0.33 (0.15, 0.72)	0.006
Any hospitalization in past 90 days	28 (57.1)	153 (68.9)	0.60 (0.32, 1.13)	0.12		
Any nursing home admission in past 90 days	9 (18.4)	65 (29.3)	0.54 (0.25, 1.18)	0.12		
Any surgery in past 90 days	24 (49.0)	74 (33.3)	1.92 (1.03, 3.59)	0.04	2.33 (1.12, 4.83)	0.024
Dialysis in past 90 days	4 (8.2)	21 (9.5)	0.85 (0.28, 2.60)	0.78		
Outpatient intravenous therapy in past 90 days	3 (6.1)	17 (7.7)	0.79 (0.22, 2.80)	0.71		
Other home therapy in past 90 days	1 (2.0)	6 (2.7)	0.75 (0.09, 6.37)	0.79		
Antimicrobials in 90 days prior to enrollment	23 (46.9)	136 (61.3)	0.56 (0.30, 1.04)	0.07		
Hospital clinic in 30 days prior to enrollment	12 (24.5)	69 (31.5)	0.71 (0.35, 1.44)	0.34		
Dialysis clinic in 30 days prior to enrollment	4 (8.2)	15 (6.8)	1.21 (0.38, 3.81)	0.75		
Chemotherapy in 30 days prior to enrollment	4 (8.2)	8 (3.7)	2.34 (0.68, 8.12)	0.18		
Systemic corticosteroids in 30 days prior to enrollment	8 (16.3)	24 (11.0)	1.59 (0.67, 3.78)	0.30		
Other immunosuppressive therapy in 30 days prior to enrollment	7 (14.3)	15 (6.8)	2.27 (0.87, 5.90)	0.09		
Radiation therapy in 30 days prior to enrollment	1 (2.0)	1 (0.5)	4.54 (0.28, 73.90)	0.29		
Urinary catheter at enrollment	5 (10.2)	86 (37.6)	0.19 (0.07, 0.49)	0.0007	0.18 (0.06, 0.51)	0.001
Vascular catheter at enrollment	15 (30.6)	56 (24.3)	1.37 (0.70, 2.70)	0.36		
Underlying disease						
Diabetes	12 (24.5)	100 (42.9)	0.43 (0.21, 0.87)	0.02	0.44 (0.20, 0.96)	0.039
Chronic pulmonary disease	12 (24.5)	49 (21.1)	1.21 (0.59, 2.50)	0.60		
Cardiovascular disease	26 (53.1)	104 (44.8)	1.39 (0.75, 2.58)	0.29		
Peripheral vascular disease	8 (16.3)	21 (9.1)	1.96 (0.81, 4.73)	0.13		
Cerebrovascular disease	5 (10.2)	50 (21.6)	0.41 (0.16, 1.10)	0.08		
Dementia	2 (4.1)	30 (12.9)	0.29 (0.07, 1.24)	0.09		
Peptic ulcer disease	0 (0.0)	6 (2.6)	0.57[a] (0.00, 4.05)	0.63		
Liver disease	4 (8.2)	19 (8.2)	1.00 (0.32, 3.07)	1.00		
Chronic renal failure	10 (20.4)	45 (19.4)	1.07 (0.49, 2.29)	0.87		
Connective tissue disease	3 (6.1)	5 (2.2)	2.96 (0.68, 12.83)	0.15	8.21 (1.49, 45.4)	0.016
HIV infection	0 (0.0)	3 (1.3)	1.23[a] (0.00, 11.56)	1.00		
Malignancy within last 5 years	14 (28.6)	36 (15.5)	2.18 (1.07, 4.45)	0.03		
Solid-organ malignancy	14 (28.6)	21 (9.1)	4.02 (1.87, 8.64)	0.0004	3.32 (1.42, 7.75)	0.006
Transplant recipient	8 (16.3)	21 (9.1)	1.96 (0.81, 4.73)	0.13		
McCabe-Jackson score D	13 (26.5)	78 (33.8)	0.71 (0.36, 1.41)	0.33		
Infection vs colonization	28 (57.1)	158 (67.8)	0.63 (0.34, 1.19)	0.15		
Source of infection				0.46		
Colonization	20 (40.8)	73 (31.5)	Baseline			
Urinary tract	10 (20.4)	82 (35.3)	0.45 (0.20, 1.01)			
Respiratory tract	5 (10.2)	19 (8.2)	0.96 (0.32, 2.89)			
Intra-abdominal	3 (6.1)	8 (3.5)	1.37 (0.33, 5.64)			
Line-related infection	3 (6.1)	7 (3.0)	1.56 (0.37, 6.60)			
Surgical site infection	2 (4.1)	5 (2.2	1.46 (0.26, 8.10)			
Primary bacteremia	1 (2.0)	13 (5.6)	0.28 (0.04, 2.28)			
Other source	5 (10.2)	25 (10.8)	0.73 (0.25, 2.15)			
Antimicrobials with Gram-negative activity 30 days prior to enrollment	23 (46.9)	125 (53.6)	0.76 (0.41, 1.42)	0.39		
Ampicillin-sulbactam	5 (10.2)	9 (3.9)	2.83 (0.90, 8.84)	0.07		
Piperacillin-tazobactam	6 (12.2)	17 (7.3)	1.77 (0.66, 4.76)	0.26		
Ceftriaxone	1 (2.0)	19 (8.2)	0.23 (0.03, 1.80)	0.16		
Cefepime	2 (4.1)	33 (14.2)	0.26 (0.06, 1.11)	0.07		
Carbapenems	2 (4.1)	11 (4.7)	0.86 (0.18, 4.00)	0.85		
Fluoroquinolones	4 (8.2)	29 (12.4)	0.63 (0.21, 1.87)	0.40		

[a] OR were estimated using median unbiased estimates.

TABLE 2 Univariate and multivariate analysis of risk factors for clinical failure at 96 h

Variable	Result by analysis type					
	Univariate				Multivariate	
	No. (%) with clinical failure ($n = 83$)	No. (%) with clinical cure ($n = 98$)	Unadjusted OR (95% CI)	*P* value	Adjusted OR (95% CI)	*P* value
Demographics						
Male gender	40 (49.4)	41 (42.3)	1.33 (0.74, 2.41)	0.34		
Age ≥65 yr	32 (38.6)	50 (51.5)	0.59 (0.33, 1.07)	0.08	0.43 (0.20, 0.95)	0.04
White race	46 (58.2)	52 (56.5)	1.07 (0.58, 1.97)	0.82		
Location of acquisition				0.002		
Community associated	11 (13.3)	7 (7.1)	Baseline			
Health care associated	35 (42.2)	67 (68.4)	0.33 (0.12, 0.93)			
Hospital acquired	37 (44.6)	24 (24.5)	0.98 (0.33, 2.88)			
Health care-associated risk factor						
Any hospitalization in past year	66 (84.6)	72 (79.1)	1.45 (0.65, 3.22)	0.36		
Any hospitalization in past 90 days	55 (69.6)	63 (67.0)	1.13 (0.59, 2.15)	0.71		
Any nursing home admission in past 90 days	11 (13.9)	23 (24.5)	0.50 (0.23, 1.10)	0.09		
Any surgery in past 90 days	42 (53.2)	35 (37.2)	1.91 (1.04, 3.52)	0.04		
Dialysis in past 90 days	12 (15.2)	5 (5.3)	3.19 (1.07, 9.48)	0.04		
Outpatient intravenous therapy in past 90 days	5 (6.3)	14 (14.9)	0.39 (0.13, 1.12)	0.08		
Other home therapy in past 90 days	3 (3.8)	2 (2.1)	1.82 (0.30, 11.15)	0.52		
Antimicrobials in 90 days prior to enrollment	49 (62.0)	64 (68.1)	0.77 (0.41, 1.43)	0.40		
Hospital clinic in 30 days prior to enrollment	21 (26.6)	45 (48.9)	0.38 (0.20, 0.72)	0.003		
Dialysis clinic in 30 days prior to enrollment	9 (11.4)	2 (2.2)	5.79 (1.21, 27.63)	0.03		
Chemotherapy in 30 days prior to enrollment	3 (3.8)	8 (8.7)	0.41 (0.11, 1.62)	0.21		
Systemic corticosteroids in 30 days prior to enrollment	10 12.7)	9 (9.8)	1.34 (0.51, 3.47)	0.55		
Other immunosuppressive therapy in 30 days prior to enrollment	10 (12.7)	8 (8.7)	1.52 (0.57, 4.07)	0.40		
Radiation therapy in 30 days prior to enrollment	0 (0.0)	1 (1.1)	1.17[a] (0.00, 45.42)	1.00		
Urinary catheter at enrollment	21 (25.9)	35 (35.7)	0.63 (0.33, 1.20)	0.16		
Vascular catheter at enrollment	32 (39.5)	24 (24.5)	2.01 (1.06, 3.82)	0.03		
Underlying disease						
Diabetes	30 (36.1)	35 (35.7)	1.02 (0.55, 1.87)	0.95		
Chronic pulmonary disease	11 (13.3)	18 (18.4)	0.68 (0.30, 1.53)	0.35		
Cardiovascular disease	43 (51.8)	31 (31.6)	2.32 (1.27, 4.26)	0.006	2.88 (1.29, 6.43)	0.01
Peripheral vascular disease	6 (7.2)	4 (4.1)	1.83 (0.50, 6.72)	0.36		
Cerebrovascular disease	13 (15.7)	20 (20.4)	0.72 (0.34, 1.56)	0.41		
Dementia	5 (6.0)	14 (14.3)	0.38 (0.13, 1.12)	0.08		
Peptic ulcer disease	1 (1.2)	2 (2.0)	0.59 (0.05, 6.57)	0.66		
Liver disease	13 (15.7)	10 (10.2)	1.63 (0.68, 3.95)	0.28		
Chronic renal failure	17 (20.5)	10 (10.2)	2.27 (0.97, 5.27)	0.06		
Connective tissue disease	2 (2.4)	1 (1.0)	2.39 (0.21, 26.89)	0.48		
HIV infection	0 (0.0)	3 (3.1)	0.30[a] (0.00, 2.84)	0.31		
Malignancy within last 5 years	17 (20.5)	21 (21.4)	0.94 (0.46, 1.94)	0.88		
Solid organ malignancy	13 (15.7)	12 (12.2)	1.33 (0.57, 3.10)	0.51		
Transplant recipient	13 (15.7)	12 (12.2)	1.33 (0.57, 3.10)	0.51		
McCabe-Jackson score D	11 (13.4)	36 (36.7)	0.27 (0.13, 0.57)	0.0006	0.39 (0.16, 0.95)	0.04
Factor associated with treatment						
Carbapenem use in 48 h after infection	12 (14.5)	24 (24.5)	0.52 (0.24, 1.12)	0.10		
Appropriate antimicrobials in first 48 h	31 (37.3)	46 (46.9)	0.67 (0.37, 1.22)	0.19		
Appropriate antimicrobials in first 72 h	38 (45.8)	66 (67.3)	0.41 (0.22, 0.75)	0.004	0.42 (0.20, 0.88)	0.02
Appropriate antimicrobials in first 96 h	52 (62.7)	77 (78.6)	0.46 (0.24, 0.88)	0.02		
Appropriate definitive therapy	61 (79.2)	87 (88.8)	0.48 (0.21, 1.11)	0.09		
Source of infection				0.0001		0.008
Urinary tract	26 (31.3)	65 (67.0)	Baseline		Baseline	
Intra-abdominal	8 (9.6)	4 (4.1)	5.00 (1.39, 18.0)		6.35 (1.51, 26.7)	
Line-related infection	3 (3.6)	6 (6.2)	1.25 (0.29, 5.38)		0.86 (0.18, 4.21)	
Primary bacteremia	6 (7.2)	7 (7.2)	2.14 (0.66, 6.98)		1.83 (0.51, 6.56)	
Respiratory tract infection	15 (18.1)	7 (7.2)	5.36 (1.96, 14.6)		2.85 (0.94, 8.66)	
Surgical site infection	5 (6.0)	3 (3.1)	4.17 (0.93, 18.7)		2.69 (0.54, 13.5)	
Other source or multiple sources	20 (24.1)	5 (5.2)	10.0 (3.39, 29.5)		8.12 (2.30, 28.6)	

[a] OR were estimated using median unbiased estimates.

TABLE 3 Distribution of resistance mechanisms in cephalosporin-resistant *Escherichia coli*

β-Lactamase	No. (%) of cephalosporin-resistant *E. coli* isolates ($n = 300$)
CTX-M	216 (72.0)
CTX-M-1 group[a]	191 (63.7)
CTX-M-2 group	2 (0.7)
CTX-M-9 group	22 (7.3)
TEM	4 (1.3)
SHV	19 (6.3)
CMY	50 (16.7)
CMY-2	32 (10.7)
CMY-22	18 (6)
KPC	7 (2.3)
FOX	1 (0.3)
Unknown/not available	9 (3)

[a] All were CTX-M-15.

group genes were identified as CTX-M-15 by the sequencing of the full structural gene. CTX-M-2 and -9 groups accounted for 0.7 and 7.3% of the isolates, respectively. The next most common group was CMY-type pAmpCs (16.7%). CMY-2 and CMY-22, which is a single-amino-acid variant of CMY-2, accounted for 64.0 and 36.0% of the CMY-type pAmpCs, respectively. In contrast, only 6.3 and 1.3% had SHV- and TEM-type ESBLs, respectively.

Antimicrobial susceptibility. MICs of various antimicrobials are shown in Table 4. Based on the CLSI 2010 breakpoints, carbapenems were the most active agents, with 95.5 and 99.0% of the isolates being susceptible to ertapenem and imipenem. Piperacillin-tazobactam, meropenem, doripenem, amikacin, nitrofurantoin, and tigecycline also were highly active, with greater than 90% of the isolates being susceptible to them. Cephalosporins were less active, with only 74.2% being susceptible to cefepime, which was still the most active cephalosporin among those tested (Tables 4 and 5). Of note, 33.2% of CTX-M producers were nonsusceptible to cefepime (Table 6). Only 17.5 and 47.1% were susceptible to ciprofloxacin and cotrimoxazole, respectively, two non-β-lactam agents commonly used for the treatment of *E. coli* infection.

DISCUSSION

We previously reported that CTX-M-15 had become the most common ESBL in *E. coli* (15), and that CMY-type pAmpC, along with ESBL, is also a common cause of cephalosporin resistance in this species at our medical center in Pittsburgh, PA (16). The aim of this study was to define this changing epidemiology in a larger, multicenter cohort of cases due to cephalosporin-resistant *E. coli* in the United States.

The study cohort consisted largely of patients with significant underlying diseases, but the 28-day mortality was relatively low at 11.1%. When we compared pAmpC- to ESBL-producing *E. coli* cases, pAmpC-producing *E. coli* isolates were significantly more frequent in patients with solid-organ malignancy, connective tissue disease, and a history of surgery in the past 90 days, while ESBL-producing *E coli* isolates were more frequent in patients with urinary catheter at enrollment, diabetes, and hospitalization

TABLE 4 Antimicrobial susceptibility of cephalosporin-resistant *Escherichia coli*[b]

	No. (%) of cephalosporin-resistant *E. coli* isolates ($n = 291$) according to:							
	CLSI 2009			CLSI 2010			MIC[a]	
Antimicrobial agent	S	I	R	S	I	R	50	90
Ampicillin				1 (0.3)	2 (0.7)	288 (99)	>16	>16
Ampicillin-sulbactam				37 (12.7)	135 (45)	119 (40.9)	16/8	>16/8
Piperacillin				16 (5.5)	46 (15.8)	229 (78.7)	>64	>64
Piperacillin-tazobactam				273 (93.8)	11 (3.8)	7 (2.4)	≤8/4	16/4
Ticarcillin-clavulanate				99 (34)	147 (50.5)	45 (15.5)	32/2	>64/2
Cefazolin	8 (2.7)	3 (1)	280 (96.2)	0	0	291 (100)	>16	>16
Ceftriaxone	50 (17.2)	56 (19.3)	185 (63.6)	15 (5.1)	9 (3.1)	267 (91.8)	>32	>32
Ceftazidime	158 (54.3)	67 (23)	66 (22.7)	80 (27.5)	78 (26.8)	133 (45.7)	8	>16
Cefepime				216 (74.2)	45 (15.5)	30 (10.3)	≤4	32
Aztreonam	83 (28.5)	61 (21)	147 (50.5)	45 (15.5)	38 (13.1)	208 (71.5)	>16	>16
Ertapenem	287 (98.6)	1 (0.3)	3 (1)	278 (95.5)	2 (0.7)	11 (3.8)	≤0.25	≤0.25
Imipenem	291 (100)	0	0	288 (99)	3 (1)	0	≤0.5	≤0.5
Meropenem	289 (99.3)	2 (0.7)	0	287 (98.6)	1 (0.3)	3 (1)	≤0.5	≤0.5
Doripenem				287 (98.6)	2 (0.7)	2 (0.7)	≤0.5	≤0.5
Amikacin				284 (97.6)	3 (1)	4 (1.4)	≤8	16
Gentamicin				169 (58.1)	8 (2.7)	114 (39.2)	≤2	>8
Tobramycin				102 (35.1)	17 (5.8)	172 (59.1)	>8	>8
Ciprofloxacin				51 (17.5)	0	240 (82.5)	>2	>2
Levofloxacin				49 (16.8)	1 (0.3)	241 (82.8)	>8	>8
Cotrimoxazole				137 (47.1)		154 (52.9)	>4/76	>4/76
Tetracycline				95 (32.6)	0	196 (67.4)	>8	>8
Minocycline				214 (73.5)	29 (10)	48 (16.5)	2	>8
Nitrofurantoin				276 (94.8)	9 (3.1)	6 (2.1)	≤32	≤32
Tigecycline				290 (99.7)	1 (0.3)	0	≤1	≤1

[a] Values are in μg/ml; 50 and 90, the MICs at which 50 and 90% of the tested isolates are inhibited, respectively. S, susceptible; I, intermediate; R, resistant.
[b] Results are based on actual MICs without adjustment for ESBL production. Isolates with unknown resistance mechanisms are not included.

TABLE 5 Cefepime MIC distributions of cephalosporin-resistant *Escherichia coli* isolates according to the β-lactamase produced

β-lactamase (*n*)	% Of isolates (cumulative %) for which the MIC (μg/ml) was:				
	Susceptible			Resistant	
	≤4	8	Intermediate (16)	32	>32
CTX-M[a] (211)	43.6 (43.6)	23.2 (66.8)	19.9 (86.7)	8.1 (94.8)	5.2 (100)
CTX-M-1 group (189)	40.2 (40.2)	23.3 (63.5)	21.7 (85.2)	9 (94.2)	5.8 (100)
CTX-M-2 group (1)	0 (0)	0 (0)	100 (100)	0 (100)	0 (100)
CTX-M-9 group (20)	80 (80)	20 (100)	0 (100)	0 (100)	0 (100)
SHV[a] (17)	94.1 (94.1)	0 (94.1)	5.9 (100)	0 (100)	0 (100)
TEM (4)	75 (75)	0 (75)	0 (75)	0 (75)	25 (100)
CMY[a] (48)	97.9 (97.9)	0 (97.9)	0 (97.9)	2.1 (100)	0 (100)
KPC (7)	71.4 (71.4)	14.3 (85.7)	14.3 (100)	0 (100)	0 (100)

[a] Coproducers were excluded from analysis.

in the past year. They represented advanced health care-associated risk factors for both groups of cases.

In the outcome analysis, early clinical success was independently associated with lower severity of disease, age of patients, cardiovascular disease, appropriate antimicrobial therapy within 72 h, and also the source of infection, primarily urinary tract infection. The presence of cardiovascular disease is not conventionally considered a risk factor for delayed clinical response in *E. coli* infection and merits further investigation. Carbapenems are the preferred antimicrobials in the treatment of serious infection due to ESBL-producing organisms (11). Carbapenem use within 48 h showed a trend for early clinical success but was not a significant factor in the multivariate analysis. However, carbapenem use at any time after infection was associated with 28-day survival in the univariate analysis. This is a potentially significant finding, given that more-seriously ill patients are likely to receive carbapenems.

In the microbiology analysis, 72.0% of cephalosporin-resistant *E. coli* isolates had CTX-M-type ESBL genes, the majority of which encoded CTX-M-15. CTX-M-15 is the most widely distributed CTX-M-type ESBL worldwide (13) and is now commonly described in the United States as well (8, 9, 12). This has an effect on the susceptibility of cefepime, as isolates producing CTX-M-15 have higher MICs of cefepime than those producing TEM- and SHV-type ESBLs (14). In the present study, the overall susceptibility to cefepime was 74.2%, whereas susceptibility to ertapenem and piperacillin-tazobactam was 95.5 and 93.8%, respectively, using the CLSI 2010 breakpoints. Cefepime thus may no longer be considered an appropriate treatment for ESBL-producing organisms where cephalosporin resistance due to CTX-M-15 is prevalent.

The recent revision of the MIC breakpoints for cephalosporins (except cefepime), aztreonam, and carbapenems by the CLSI (3) affected ceftazidime and aztreonam the most in our study, for which the overall susceptibility dropped from 54.3 to 27.5% and from 28.5 to 15.5% under the new breakpoints, respectively. Even with this revision, however, a substantial portion of ESBL-producing *E. coli* isolates remained susceptible to cephalosporins and aztreonam and would be missed in the absence of the phenotypic confirmation of ESBL production, which is now recommended only as an optional procedure (3). This finding was in agreement with a recent study from China (18). CMY-producing *E. coli*, on the other hand, would be consistently reported as nonsusceptible to cephalosporins and also to aztreonam in most instances under the revised breakpoints.

Our study has several limitations. Since the study was observational, the assessment of clinical outcome was based on information available from the medical records. Also, we elected to compare the pAmpC and ESBL cases for the risk factor analysis to determine risk factors that are unique to pAmpC cases. This approach may have missed factors that otherwise would have been significant when using fully susceptible cases as controls.

In summary, this study, which is the largest that has been con-

TABLE 6 Susceptibility of cephalosporin-resistant *Escherichia coli* to ceftazidime, ceftriaxone, cefepime, and aztreonam using the CLSI 2009 and CLSI 2010 breakpoints according to the β-lactamase produced

β-lactamase type (*n*)	% Of isolates susceptible to:							
	Ceftazidime		Ceftriaxone		Aztreonam		Ertapenem	
	CLSI 2009	CLSI 2010	CLSI 2009	CLSI 2010	CLSI 2009	CLSI 2010	CLSI 2009	CLSI 2010
CTX-M[a] (211)	62.6	32.2	8.5	1.4	22.7	15.2	99.5	98.1
CTX-M-1 (189)	58.7	25.4	7.9	1.6	14.7	7.4	99.5	97.9
CTX-M-2 (1)	100	100	0	0	0	0	100	100
CTX-M-9 (20)	100	95	15	0	95	80	100	100
SHV[a] (17)	82.3	47	88.2	52.9	41.1	23.5	100	100
TEM (4)	25	0	100	75	75	50	75	75
CMY[a] (48)	12.5	0	14.6	0	47.9	12.5	97.9	93.8
KPC (7)	42.9	28.6	71.4	0	0	0	85.7	28.6

[a] Coproducers were excluded.

ducted on this topic in the United States, reveals the contemporary epidemiology of cephalosporin resistance in *E. coli*, which is now dominated by CTX-M-15 ESBL and, to a lesser extent, by CMY-type pAmpC. This shift in epidemiology has compromised the activity of cefepime, whereas the activity of carbapenems and piperacillin-tazobactam are maintained among β-lactams. These findings have implications in the selection of appropriate empirical therapy when infection due to *E. coli* is suspected in settings where resistance to cephalosporin is common.

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