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Community-onset extended-spectrum-β-lactamase-producing *Escherichia coli* sequence type 131 at two Korean community hospitals: The spread of multidrug-resistant *E. coli* to the community via healthcare facilities

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ABSTRACT

Background: The recent molecular epidemiology of ESBL-producing *Escherichia coli* infection in two Korean community hospitals was evaluated in this prospective observational study.
Methods: We collected non-duplicated *E. coli* isolates from consecutive, sequentially encountered patients with community-onset episodes between March and April 2016 in two community hospitals in Gyeonggi-do province, Korea. We studied the prevalence, clinical characteristics and molecular epidemiology of *E. coli* sequence type 131 (ST131) isolated from the community.
Results: From a total of 213 *E. coli* isolates collected from the community, 94 (44.1%) were community-onset healthcare-associated isolates and 119 (55.9%) were community-associated isolates, of which urinary tract infection was the majority. A total of 55 (25.8%) of the 213 *E. coli* isolates were confirmed to have ESBL genes, which were mainly CTX-M types such as CTX-M-14 and CTX-M-15. There was no difference in the proportion of globally epidemic ST131 clones or that of O25, O16, *H30*, or *H30Rx* subclones between community-associated and community-onset healthcare-associated isolates.
Conclusions: In this study, considerable ST131 *E. coli* isolations in the community were observed and about half of them were related to the history of a visit to the healthcare facilities, indicating the spread of multidrug-resistant *E. coli* to the community via healthcare facilities.

1. Introduction

Extraintestinal pathogenic *Escherichia coli* (ExPEC) infection is a significant public health problem, consisting primarily of urinary tract infections.[1] Of ExPEC infections, sequence type 131 (ST131) *E. coli* has played a major part in the global dissemination of specific traits for epidemic expansion such as greater virulence and multidrug resistance, including extended-spectrum β-lactamase (ESBL) and fluoroquinolone resistance.[2] The well-defined characteristics of this clone consist of phylogenetic B2, serotype O25, *fimH* type *H30* and ST131 according to multilocus sequence typing (MLST) based on Achtman.[3] Especially, single expansion of the ST131 *E. coli* subclone, *H30Rx* is associated with fluoroquinolone resistance and CTX-M-15 ESBL and comprises most ST131 clones.[4] Another ST131 *E. coli* subclone was recently reported, namely, the serogroup O16, which shows resistance to ampicillin, gentamicin, and trimethoprim-sulfamethoxazole and is susceptible to fluoroquinolones and extended-spectrum cephalosporins.[5]

The aims of this study were to evaluate the recent molecular epidemiology of ESBL-producing *E. coli* acquisitions from two community hospitals, and to assess the differences between community-associated and community-onset healthcare-associated isolates in a prospective, multicenter, observational study.[6]

2. Materials and methods

2.1. Bacterial isolates

We collected non-duplicated *E. coli* isolates from consecutive, sequentially encountered patients with community-onset

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episodes in outpatient departments or within 48 hours of hospitalization between March and April 2016 in two community hospitals in Gyeonggi-do province, Korea (742 beds, Goyang-si; 132 beds, Yongin-si). Healthcare-acquired *E. coli* isolates that were collected after 48 hours of admission were excluded. The study was approved by institutional review boards as required by local hospital policy (NHIMC 2016-02-006).

2.2. Definitions

Sites of acquisition (community-associated or healthcare-associated) were determined as described by Friedman with some modifications.[7,8] Healthcare-associated infections have any of following histories: attended a hospital or hemodialysis clinic or received intravenous chemotherapy in the 30 days before the infection; hospitalized in an acute care hospital for 2 or more days in the 90 days before the infection. Others were defined as community-associated infection. The diagnosis of infection was made based on clinical, bacteriological, and radiological investigations.[9]

2.3. Microbiological analysis

Identification and susceptibility testing were performed using the Vitek 2 automated analyzer system (bioMérieux, Marcy l'E'toile, France) with a VITEK AST2 N212 card (bioMérieux). Antibiotic susceptibility was interpreted using CLSI criteria.[10] ESBL genotype was determined by PCR and sequencing.[11] For the detection of ST131, all isolates were screened by PCR for O16-ST131 and O25-ST131.[5] *FimH* type and *H30Rx* were determined by PCR and sequencing.[4,12]

2.4. Statistical analysis

SAS software, version 9.2 was used for the statistical analysis (SAS Institute, Cary, NC, USA). Comparisons of proportions were tested using the Chi-square test or Fisher's exact test with the level of significance set as P value < 0.05.

3. Results

A total of 213 community-onset isolates were identified, consisting of 94 (44.1%) healthcare-associated (HA) isolates and 119 (55.9%) community-associated (CA) isolates, the majority of which were from urinary tract infections. No statistically significant difference was found in either infection or colonization status of CA isolates, compared to that of HA isolates ($P = 0.701$, Table 1). Although urinary tract infection was the most common type in both groups, bacteremia was more frequently encountered in the HA group than in the CA group and the two groups were different in the type of infection ($P < 0.0001$, Table 1). A total of 55 *E. coli* isolates (25.8%, 55/213) had ESBL genes, which were mainly CTX-M types such as CTX-M-15 and CTX-M-14. In addition, there was no statistically significant difference in ESBL genotypes between the CA and HA groups ($P = 0.8752$, Table 1).

Among CA isolates, 26.9% (32/119) were caused by the globally epidemic ST131 strain, which was similar to that of HA isolates (27.7%, 26/94) and there was no statistically significant difference in the proportion of globally epidemic ST131 clone or O25, O16, *H30*, or *H30Rx* subclones between CA and HA isolates (Table 1).

According to ST131 subclones, ESBL positive rates and types were different (Table 2). ST131-O25*H30Rx* subclones most commonly produced ESBLs, all of which were CTX-M-15 types,

Table 1
Types of infections, extended-spectrum β-lactamase (ESBL) types, and the prevalence of sequence type 131 (ST131) of community-onset healthcare-associated (HA) and community-associated (CA) *Escherichia coli* episodes (N = 213)

	HA (N = 94)	CA (N = 119)	*P*
Status of infection			0.7016
Infection	74 (78.7%)	98 (82.4%)	
Colonization	17 (18.1%)	19 (16.0%)	
Indeterminate	3 (3.1%)	2 (1.7%)	
Type of infection			<0.0001
Urinary tract infection	46 (62.2%)	78 (79.6%)	
Bacteremia	18 (24.3%)	9 (9.2%)	
Abscess/wound	8 (10.8%)	1 (1.0%)	
Others	2 (2.7%)	10 (10.2%)	
ESBL by genotype	25 (26.6%)	30 (25.2%)	0.8752
CTX-M group	CTX-M-15 (N = 9), CTX-M-14 (N = 10), CTX-M-27 (N = 5), other (N = 1)	CTX-M-15 (N = 17), CTX-M-14 (N = 9), CTX-M-27 (N = 1), other (N = 3)	
SHV group	Not detected	Not detected	
TEM group	Not detected, TEM-1[a] (N = 2)	Not detected, TEM-1[a] (N = 1)	
ST131 subclone	26 (27.7%)	32 (26.9%)	1.0000
O16-ST131	10 (10.6%)	7 (5.9%)	0.2152
O25-ST131	16 (17.1%)	25 (21%)	0.4893
H30	15/16 (93.8%)	20/25 (80%)	0.3759
H30Rx	5/15 (33.3%)	11/20 (55%)	0.3064
Non-ST131	68 (72.3%)	87 (73.1%)	1.0000

N, number; Chi-square test or Fisher's exact test.
[a] Narrow-spectrum β-Lactamase.

Table 2
Extended-spectrum β-lactamase (ESBL) types and the prevalence of sequence type 131 (ST131) in community-onset *Escherichia coli* episodes (N = 58)

	ST131 (O25+O16)	O25	*H30*	*H30Rx*	O16
ESBL positive (%)	28/58 (48.3%)	23/41 (56.1%)	20/35 (57.1%)	13/16 (81.3%)	5/17 (29.4%)
CTX-M group					
CTX-M-15	16	16	14	13	0
CTX-M-14	9	4	4	0	5
CTX-M-27	3	3	2	0	0

N, number.

Table 3
Antimicrobial resistance rate (%) of *Escherichia coli* isolated from two community hospitals

Antimicrobial agents	Resistance rate (%)						
	Site of acquisition		Sequence type (ST)				
	HA[a] (n = 93)	CA (n = 119)	ST131-O25-*H30* (n = 35)	ST131-O25-*H30Rx* (n = 16)	ST131-O16 (n = 17)	All ST131 (n = 58)	Other STs (n = 154)
Ampicillin	70	68	83	94	65	79	65
Piperacillin	69	64	80	88	65	78	62
Ampicillin/sulbactam	33	25	40	44	29	35	27
Cefoxitin	4	2	3	6	0	3	3
Cefotaxime	30	27	60	88	29	50	20
Ceftazidime	30	24	57	81	24	47	19
Meropenem	0	0	0	0	0	0	0
Imipenem	0	0	0	0	0	0	0
Ciprofloxacin	39	44	100	100	18	67	32
Gentamicin	23	25	51	56	35	43	17
Tobramycin	13	11	31	38	24	28	6
Amikacin	0	0	0	0	0	0	0
Cotrimoxazole	33	25	49	44	41	45	23

HA, community-onset healthcare-associated; CA, community-associated; all ST131 include O25-*H30*, O25-*nonH30*, and O16.
[a] One isolate was excluded from the antimicrobial susceptibility test.

whereas only 29.4% of ST131-O16 subclones produced ESBLs, all of which were CTX-14 types.

Antimicrobial resistance patterns showed distinct characteristics according to ST131 subclone, and in particular the *H30Rx* group showed prominent resistance to fluoroquinolones such as all resistant to ciprofloxacin or levofloxacin and 88% resistance to cefotaxime due to CTX-M type ESBL production (Table 3). However, the antimicrobial resistance rates were similar in the HA and CA groups (Table 3).

4. Discussion

It is well known that the majority of community-onset *E. coli* isolates come from urinary tract infections,[13] and our study also showed that urinary tract infection was the most common type. Recently, the emergence of community-onset bacteremia via ESBL-producing *E. coli* has become a major concern.[8,14] In this study, community-onset bacteremia was more frequently encountered in the HA group (possibly not as healthy as the CA group), suggesting that not only microbiological factors but also the impact of host factors are important for severe types of infections such as bacteremia.

Extended-spectrum β-lactamase (ESBL)-producing *E. coli* has become widespread in hospitals around the world since the late 1980s,[15] but a sudden worldwide increase in the mid 2000s is mainly due to sequence type (ST) 131 with resistance to quinolone and 3rd generation cephalosporin, which was suggested as the main cause of the spread of ESBL producing *E. coli* in the community.[3,16] Not only community-onset *E. coli* infection but also asymptomatic carriage among health individuals without prior exposure to antibiotics increased.[17] Our study showed a high prevalence of ESBLs, mainly CTX-M, among community-onset *E. coli* isolates in Korean community hospitals. In the Asia-Pacific region, higher rates of ESBL-producing *E. coli* were observed in countries like China, India and Thailand, compared to Korea.[18]

The incidence of notorious epidemic resistant clones of *E. coli* varies over time and geographic regions, and the ST131 *E. coli* clonal group has been evolving through the acquisition of new mobile elements.[19] Continuous monitoring of major epidemic resistance clones is needed for updated information regarding the status of ST 131 *E. coli*. In the most recent molecular epidemiology data of ST131 *E. coli* in Korea, the ST131 clonal group comprised 21% (57/268) of all *E. coli* isolates, 37% (21/57) of which were *H30Rx* subclones.[20] This study included both healthcare-onset and community-onset *E. coli* isolates, collected from urine or blood cultures between September 2012 and January 2013. Although direct comparisons are inadequate, considering different acquisition sites (our study includes only *E. coli* isolates from the community), our study showed that 27% (58/213) of *E. coli* isolates from the community belonged to the globally epidemic ST131 strain, 28% (16/58) of which were *H30Rx* subclones. Therefore ST131 *E. coli* infections in the community are already considerable in Korea, and ST131-O25*H30Rx* subclones could be considered potential driving factors, with their high ESBL (mainly CTX-M type) production rates as well as successful traits related to extensive virulence gene content and ongoing transmission.[2]

Recently multiple sources of ST131 *E. coli* in the community have been suspected, including livestock, companion animals, sewage, waste water, and recreational waterways.[21,22] This 'One Health' concept, which recognizes that the health of humans is connected to the health of animals and the environment, may be useful for promoting cross-sectorial collaborations and represents a possible solution for preventing the spread of multidrug resistance in the community. Nevertheless, it is important to emphasize that ST131 *E. coli* accounts for a large proportion of HA infections.[23] In our study, 44.1% (94/213) of patients had HA factors such as recent visits to a healthcare facility. Thus, It is reasonable to assume that the burden of ESBL producing *E. coli* in the community is affected by healthcare facilities (and vice versa) in a 'revolving door' pattern,[24] similar to that of CA methicillin-resistant *Staphylococcus aureus*.[25]

In this study, considerable community-onset acquisitions of ST131 *E. coli* from two community hospitals were observed, and about half of them had history related to healthcare facilities, indicating the spread of multidrug-resistant *E. coli* to the community via healthcare facilities.

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Conflict of Interest: None.

References

1. Vila J, Sáez-López E, Johnson JR, Römling U, Dobrindt U, Cantón R, et al. *Escherichia coli*: an old friend with new tidings. *FEMS Microbiol Rev* 2016;**40**:437–63.

2. Colpan A, Johnston B, Porter S, Clabots C, Anway R, Thao L, et al. *Escherichia coli* sequence type 131 (ST131) subclone *H30* as an emergent multidrug-resistant pathogen among US veterans. *Clin Infect Dis* 2013;**57**:1256–65.
3. Johnson JR, Urban C, Weissman SJ, Jorgensen JH, Lewis 2nd JS, Hansen G, et al. Molecular epidemiological analysis of *Escherichia coli* sequence type ST131 (O25:H4) and blaCTX-M-15 among extended-spectrum-β-lactamase-producing *E. coli* from the United States, 2000 to 2009. *Antimicrob Agents Chemother* 2012;**56**:2364–70.
4. Price LB, Johnson JR, Aziz M, Clabots C, Johnston B, Tchesnokova V, et al. The epidemic of extended-spectrum-β-lactamase-producing *Escherichia* coli ST131 is driven by a single highly pathogenic subclone, *H30-Rx*. *mBio* 2013;**17**:1–10.
5. Johnson JR, Clermont O, Johnston B, Clabots C, Tchesnokova V, Sokurenko E, et al. Rapid and specific detection, molecular epidemiology, and experimental virulence of the O16 subgroup within *Escherichia coli* Sequence Type 131. *J Clin Microbiol* 2014;**52**:1358–65.
6. Cheong HS, Kang CI, Kwon KT, Heo ST, Wi YM, Kim ES, et al. Clinical significance of healthcare-associated infections in community-onset *Escherichia coli* bacteraemia. *J Antimicrob Chemother* 2007;**60**:1355–60.
7. Friedman ND, Kaye KS, Stout JE, McGarry SA, Trivette SL, Briggs JP, et al. Health care-associated bloodstream infections in adults: a reason to change the accepted definition of community-acquired infections. *Ann Intern Med* 2002;**137**:791–7.
8. Park YS, Bae IK, Kim J, Jeong SH, Hwang S, Seo Y, et al. Risk factors and molecular epidemiology of community-onset extended-spectrum β-Lactamase-producing *Escherichia coli* bacteremia. *Yonsei Med J* 2014;**55**:467–75.
9. Tang HJ, Hsieh CF, Chang PC, Chen JJ, Lin YH, Lai CC, et al. Clinical significance of community- and healthcare-acquired carbapenem-resistant *Enterobacteriaceae* isolates. *PLoS One* 2016;**11**:e0151897.
10. Performance Standards for Antimicrobial Susceptibility Testing: 26th Informational Supplement. CLSI document M100S (ISBN 1-56238-923-8). 2016, Wayne, PA, USA: Clinical and Laboratory Standards Institute.
11. Sidjabat HE, Paterson DL, Adams-Haduch JM, Ewan L, Pasculle AW, Muto CA, et al. Molecular epidemiology of CTX-M-producing *Escherichia coli* isolates at a tertiary medical center in western Pennsylvania. *Antimicrob Agents Chemother* 2009;**53**:4733–9.
12. Weissman SJ, Johnson JR, Tchesnokova V, Billig M, Dykhuizen D, Riddell K, et al. High-resolution two-locus clonal typing of extraintestinal pathogenic *Escherichia coli*. *Appl Environ Microbiol* 2012;**78**:1353–60.
13. Doi Y, Park YS, Rivera JI, Adams-Haduch JM, Hingwe A, Sordillo EM, et al. Community-associated extended-spectrum β-lactamase-producing *Escherichia coli* infection in the United States. *Clin Infect Dis* 2013;**56**:641–8.
14. Cha MK, Kang C, Kim SH, Cho SY, Ha YE, Wi YM, et al. Comparison of the microbiological characteristics and virulence factors of ST131 and non-ST131 clones among extended-spectrum β-lactamase–producing *Escherichia coli* causing bacteremia. *Diagn Microbiol and Infect Dis* 2016;**84**:102–4.
15. Paterson DL, Bonomo RA. Extended-spectrum β-lactamases: a clinical update. *Clin Microbiol Rev* 2005;**18**:657–86.
16. Ben-Ami R, Rodríguez-Baño J, Arslan H, Pitout JD, Quentin C, Calbo ES, et al. A multinational survey of risk factors for infection with extended-spectrum beta-lactamase-producing *Enterobacteriaceae* in nonhospitalized patients. *Clin Infect Dis* 2009;**49**:682–90.
17. Spellberg B, Doi Y. The Rise of Fluoroquinolone-Resistant *Escherichia coli* in the Community: Scarier Than We Thought. *J Infect Dis* 2015;**212**:1853–5.
18. Sheng WH, Badal RE, Hsueh PR. SMART Program. Distribution of extended-spectrum β-lactamases, AmpC β-lactamases, and carbapenemases among *Enterobacteriaceae* isolates causing intra-abdominal infections in the Asia-Pacific region: results of the study for Monitoring Antimicrobial Resistance Trends (SMART). *Antimicrob Agents Chemother* 2013;**57**:2981–8.
19. Zakour NLB, Alsheikh-Hussain AS, Ashcroft MM, Nhu NTK, Roberts LW, Stanton-Cook M, et al. Sequential acquisition of virulence and fluoroquinolone resistance has shaped the evolution of *Escherichia coli* ST131. *mBio* 2016;**7**:347–416.
20. Kim S, Park Y, Johnson JR, Yu JK, Kim Y, Kim YS. Prevalence and characteristics of *Escherichia coli* sequence type 131 and its *H30* and *H30Rx* subclones: a multicenter study from Korea. *Diagn Microbiol Infect Dis* 2016;**84**:97–101.
21. Lazarus B, Paterson DL, Mollinger JL, Rogers BA. Do human extraintestinal *Escherichia coli* infections resistant to expanded-spectrum cephalosporins originate from food-producing animals? A systematic review. *Clin Infect Dis* 2015;**60**:439–52.
22. Colomer-Lluch M, Mora A, López C, Mamani R, Dahbi G, Marzoa J, et al. Detection of quinolone-resistant *Escherichia coli* isolates belonging to clonal groups O25b:H4-B2-ST131 and O25b:H4-D-ST69 in raw sewage and river water in Barcelona, Spain. *J Antimicrob Chemother* 2013;**68**:758–65.
23. Rogers BA, Doi Y. Who is leading the dance? Understanding the spread of *Escherichia coli* sequence type 131. *Infect Control Hosp Epidemiol* 2013;**34**:370–2.
24. Burke L, Humphreys H, Fitzgerald-Hughes D. The revolving door between hospital and community: extended-spectrum beta-lactamase-producing *Escherichia coli* in Dublin. *J Hosp Infect* 2012;**81**:192–8.
25. Seybold U, Kourbatova EV, Johnson JG, Halvosa SJ, Wang YF, King MD, et al. Emergence of community-associated methicillin-resistant *Staphylococcus aureus* USA300 genotype as a major cause of health care-associated blood stream infections. *Clin Infect Dis* 2006;**42**:647–56.