

Genomic Epidemiology of Global Carbapenemase-Producing *Escherichia coli*, 2015–2017

Gisele Peirano, Liang Chen, Diego Nobrega, Thomas J. Finn, Barry N. Kreiswirth, Rebekah DeVinney, Johann D.D. Pitout



In support of improving patient care, this activity has been planned and implemented by Medscape, LLC and Emerging Infectious Diseases. Medscape, LLC is jointly accredited by the Accreditation Council for Continuing Medical Education (ACCME), the Accreditation Council for Pharmacy Education (ACPE), and the American Nurses Credentialing Center (ANCC), to provide continuing education for the healthcare team.

Medscape, LLC designates this Journal-based CME activity for a maximum of 1.00 **AMA PRA Category 1 Credit(s)**[™]. Physicians should claim only the credit commensurate with the extent of their participation in the activity.

Successful completion of this CME activity, which includes participation in the evaluation component, enables the participant to earn up to 1.0 MOC points in the American Board of Internal Medicine's (ABIM) Maintenance of Certification (MOC) program. Participants will earn MOC points equivalent to the amount of CME credits claimed for the activity. It is the CME activity provider's responsibility to submit participant completion information to ACCME for the purpose of granting ABIM MOC credit.

All other clinicians completing this activity will be issued a certificate of participation. To participate in this journal CME activity: (1) review the learning objectives and author disclosures; (2) study the education content; (3) take the post-test with a 75% minimum passing score and complete the evaluation at <http://www.medscape.org/journal/eid>; and (4) view/print certificate. For CME questions, see page XXX.

Release date: April 13, 2022; Expiration date: April 13, 2023

Learning Objectives

Upon completion of this activity, participants will be able to:

- Assess the global distribution of different carbapenemase genes, based on a genome sequencing study of 229 carbapenemase-producing *Escherichia coli* (2015–17) from 36 countries
- Evaluate antimicrobial resistance determinants and plasmid replicon types, virulence-associated factors, and carbapenemase gene flanking regions and plasmid analysis, based on a genome sequencing study of 229 carbapenemase-producing *Escherichia coli* (2015–17) from 36 countries
- Determine the public health implications of the global distribution of different carbapenemase genes and associated factors, based on a genome sequencing study of 229 carbapenemase-producing *Escherichia coli* (2015–17) from 36 countries

CME Editor

Jude Rutledge, BA, Technical Writer/Editor, Emerging Infectious Diseases. *Disclosure: Jude Rutledge has disclosed no relevant financial relationships.*

CME Author

Laurie Barclay, MD, freelance writer and reviewer, Medscape, LLC. *Disclosure: Laurie Barclay, MD, has disclosed the following relevant financial relationships: stocks, stock options, or bonds from AbbVie (former).*

Authors

Gisele Peirano, PhD; Liang Chen, PhD; Diego Nobrega, PhD; Thomas J. Finn, PhD; Barry N. Kreiswirth, PhD; Rebekah DeVinney, PhD; and Johann D.D. Pitout, MD.

Author affiliations: University of Calgary, Calgary, Alberta, Canada (G. Peirano, T.J. Finn, R. DeVinney, J.D.D. Pitout); Alberta Precision Laboratories, Calgary (G. Peirano, J.D.D. Pitout); Hackensack Meridian School of Medicine,

Nutley, New Jersey, USA (L. Chen, B.N. Kreiswirth); University of Guelph, Guelph, Ontario, Canada (D. Nobrega); University of Pretoria, Pretoria, South Africa (J.D.D. Pitout)

DOI: <https://doi.org/10.3201/eid2805.212535>

We describe the global molecular epidemiology of 229 carbapenemase-producing *Escherichia coli* in 36 countries during 2015–2017. Common carbapenemases were oxacillinase (OXA) 181 (23%), New Delhi metallo-β-lactamase (NDM) 5 (20%), OXA-48 (17%), *Klebsiella pneumoniae* carbapenemase 2 (15%), and NDM-1 (10%). We identified 5 dominant sequence types (STs); 4 were global (ST410, ST131, ST167, and ST405), and 1 (ST1284) was limited to Turkey. OXA-181 was frequent in Jordan (because of the ST410-B4/H24RxC subclade) and Turkey (because of ST1284). We found nearly identical IncX3-*bla*_{OXA-181} plasmids among 11 STs from 12 countries. NDM-5 was frequent in Egypt, Thailand (linked with ST410-B4/H24RxC and ST167-B subclades), and Vietnam (because of ST448). OXA-48 was common in Turkey (linked with ST11260). Global *K. pneumoniae* carbapenemases were linked with ST131 C1/H30 subclade and NDM-1 with various STs. The global carbapenemase *E. coli* population is dominated by diverse STs with different characteristics and varied geographic distributions, requiring ongoing genomic surveillance.

Carbapenems are effective options available for treating serious infections caused by multidrug-resistant (MDR) Enterobacterales bacteria (1). The emergence of carbapenem resistance is a major public health concern, and the World Health Organization has identified carbapenem-resistant Enterobacterales as critical-priority bacteria (2).

Carbapenemases are important causes of carbapenem resistance (3). Carbapenemase genes can be transferred between Enterobacterales species. The most common carbapenemases among Enterobacterales are *Klebsiella pneumoniae* carbapenemases (KPCs), imipenemases (IMPs), Verona integron-encoded metallo-β-lactamases (VIMs), New Delhi metallo-β-lactamases (NDMs), and oxacillinase (OXA) 48-like enzymes. *Escherichia coli* is the second most common carbapenemase-producing Enterobacterales species (4,5).

Because *E. coli* is mainly responsible for human community-associated infections (6), it evades conventional hospital-based infection-prevention measures (7). *E. coli* is an important One Health (i.e., human, animal, environmental health) reservoir for antimicrobial resistance (AMR) genes (8). Tracking global mobile genetic elements and *E. coli* clones associated with carbapenemase genes is a public health priority (9) and aids in designing management and prevention strategies.

Comprehensive epidemiology data about carbapenemase-producing *E. coli* is limited to institutional, regional, or countrywide surveys (10). We used short-read whole-genome sequencing (WGS) to

describe the molecular characteristics and international distribution of carbapenemase-producing *E. coli*. We describe the geographic distribution of different carbapenemase genes (including their associations with dominant sequence types [STs], clades and underlying mobile genetic elements), other β-lactamases, AMR genes, and virulence factors.

Materials and Methods

Bacterial Isolates

We obtained ethics approval for this study through the University of Calgary Conjoint Health Research Ethics Board (approval no. REB17-1010). We included 229 clinical, nonrepeat *E. coli* isolates collected from 2 global surveillance programs (SMART and INFORM) during 2015–2017 (Appendix, <https://wwwnc.cdc.gov/EID/article/28/5/21-2535-App1.pdf>). Isolates had undergone identification and susceptibility testing using Clinical Laboratory and Standards Institute guidelines (4,5,11). Carbapenem nonsusceptible isolates underwent molecular screening for *bla*_{KPC}, *bla*_{VIM}, *bla*_{NDM}, *bla*_{OXA-48-like}, *bla*_{IMP}, and *bla*_{GES}, as described previously (4,5). Overall, we collected 87,182 Enterobacterales for the period 2015–2017 from 62 countries: 27,444 were identified as *E. coli* and 275 (1%) tested nonsusceptible to ≥1 of the carbapenems. Most (229 [83%]) were positive for either *bla*_{KPC}, *bla*_{OXA-48-like}, *bla*_{NDM}, *bla*_{VIM}, or *bla*_{IMP} and were included in this study. The remaining 46 were negative for *bla*_{KPC}, *bla*_{VIM}, *bla*_{NDM}, *bla*_{OXA-48-like}, *bla*_{IMP}, and *bla*_{GES}.

We defined major STs as representing >10% and minor STs as representing 5%–10% of the total *E. coli* carbapenemase population (12). Dominant STs were both major and minor STs.

Genomic Analysis

We subjected the carbapenemase-producing *E. coli* (n = 229) to short-read WGS by using NovoSeq (Illumina, <https://www.illumina.com>) with 151 × 2 paired-end reads (13,14). We obtained draft genomes by using SPAdes 3.15 (15). We used BLAST (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>) to determine AMR genes, plasmid replicons, and virulence genes against the following databases or typing schemes: National Center for Biotechnology Information Bacterial Antimicrobial Resistance Reference Gene Database (<https://www.ncbi.nlm.nih.gov/bioproject/PRJNA313047>), ResFinder (16), PlasmidFinder (17), multilocus sequence typing (18), and virulence finder (19). We conducted multilocus sequence typing by using *mlst* 2.19 (<https://github.com/tseemann/mlst>). We identified ST410 and ST131 clades and subclades as described previously (20,21).

RESEARCH

For phylogenetic analyses, we mapped trimmed raw reads from each genome to a reference genome sequence (EC958 [GenBank accession no. HG941718] for ST131, JS316 [GenBank accession no. CP058618] for ST410, WCHEC005237 [GenBank accession no. CP026580] for ST167, and AR_0015 [GenBank accession no. CP024862] for ST405) by using snippy (<https://github.com/tseemann/snippy>). We filtered single-nucleotide polymorphisms (SNPs) among prophages, repeated sequences, or insertion sequences as previously described (22), and we generated a maximum-likelihood phylogenetic tree inferred from the resulting SNP alignment by using RAxML 8.2.12 by using a general time-reversible model of nucleotide substitution and 4 discrete γ categories of rate heterogeneity (23). We identified phylogenetic clades by

using hierarchical Bayesian analysis of the population structure in R by using RhierBAPS with 10 initial clusters at 2 clustering levels (24). We defined clades by using the first level of clustering and subclades at the second level of clustering (25). We annotated the phylogenetic trees in iTOL (26). We deposited all sequencing data in the National Center for Biotechnology Information database (BioProject PRJNA780590).

Statistical Analyses

We conducted all analyses in R 3.6.1 (27). Initially, we attempted to fit generalized linear mixed models with country-level random effects to summarize comparisons between dominant STs with respect to antimicrobial and virulence genes. Most models failed to converge, possibly because of the low number of isolates

Table. Global molecular epidemiology of 229 carbapenemase-producing *Escherichia coli* isolates, 36 countries, 2015–2017*

Carbapenemases (no. isolates)	Geographic location (no. isolates)	Sequence types (no. isolates)
KPCs (50)		
KPC-2 (35)	Argentina (4), Brazil (5), Colombia (8), Greece (1), Guatemala (4), Israel (2), Puerto Rico (2), United States (4), Venezuela (1), Vietnam (4)	ST10 (3), ST46 (2), ST69 (2), ST95 (3), ST131 (7), ST349 (1), ST405 (3), ST410 (3), ST538 (1), ST540 (1), ST607 (1), ST617 (1), ST648 (1), ST1193 (1), ST1196 (1), ST2172 (1), ST2279 (1), ST3580 (1)
KPC-3 (14)	Colombia (1), Israel (1), Italy (8), United States (4)	ST12 (1), ST73 (1), ST131 (7), ST141 (1), ST191 (1), ST617 (1), ST973 (1), ST1148 (1)
KPC-18 (1)	United States (1)	ST131 (1)
NDMs (66)		
NDM-1 (19)	Egypt (3), Guatemala (2), Kuwait (1), Morocco (4), Philippines (1), Romania (1), Russia (3), Serbia (1), Thailand (2), Vietnam (1)	ST38 (1), ST44 (1), ST69 (1), ST95 (1), ST131 (4), ST167 (3), ST345 (1), ST361 (1), ST617 (2), ST1193 (1), ST1434 (1), ST1470 (1), ST4553 (1)
NDM-4 (1)	Vietnam (1)	ST405 (1)
NDM-5 (40)	Canada (1), Egypt (16), Italy (2), Jordan (4), Lebanon (1), Thailand (8), United Kingdom (2), Vietnam (6)	ST131 (1), ST156 (1), ST167 (11), ST361 (4), ST405 (3), ST410 (12), ST448 (2), ST648 (4), ST2003 (2)
NDM-6 (1)	Guatemala (1)	ST38 (1)
NDM-7 (5)	Philippines (4), Vietnam (1)	ST156 (2), ST410 (1), ST448 (1), ST5229 (1)
OXA-48-like (96)		
OXA-48 (40)	Austria (1), Belgium (2), Egypt (3), Georgia (3), Israel (1), Lebanon (2), Mexico (1), Morocco (2), Saudi Arabia (1), Spain (2), Thailand (1), Tunisia (1), Turkey (15), United Kingdom (1), Vietnam (4)	ST10 (2), ST12 (1), ST34 (1), ST38 (8), ST58 (1), ST131 (2), ST224 (1), ST349 (1), ST354 (6), ST361 (1), ST405 (4), ST410 (2), ST624 (1), ST648 (1), ST1431 (1), ST11260 (6)
OXA-181 (48)	Egypt (6), Germany (1), Jordan (15), Kuwait (1), Lebanon (1), Malaysia (1), South Africa (2), Taiwan (1), Thailand (2), Turkey (18)	ST46 (1), ST131 (1), ST167 (2), ST205 (1), ST354 (1), ST410 (21), ST648 (1), ST1284 (18), ST1487 (1), ST6802 (1)
OXA-232 (5)	Malaysia (1), Mexico (3), Thailand (1)	ST127 (1), ST131 (1), ST361 (3)
OXA-244 (3)	Egypt (3)	ST58 (1), ST648 (1), ST1722 (1)
VIMs (4)		
VIM-1 (2)	Greece (1), Spain (1)	ST88 (1), ST404 (1)
VIM-23 (2)	Mexico (2)	ST410 (2)
IMPs (2)		
IMP-59 (2)	Australia (2)	ST357 (2)
Two carbapenemases (11)		
NDM-1 + VIM-1 (1)	Egypt (1)	ST131 (1)
NDM-1 + OXA-181 (2)	Egypt (2)	ST46 (2)
NDM-5 + OXA-48 (1)	Egypt (1)	ST167 (1)
NDM-5 + OXA-181 (5)	Egypt (3), South Korea (1), Vietnam (1)	ST410 (4), ST448 (1)
NDM-5 + OXA-232 (2)	United Kingdom (2)	ST2083 (2)

*KPC, *Klebsiella pneumoniae* carbapenemase; NDM, New Delhi metallo- β -lactamase; OXA, oxacillinase; ST, sequence type; VIM, Verona integron-encoded metallo- β -lactamase.

for some STs and the large number of countries involved. Thereafter, we attempted to use exact logistic regression models for clustered data, as previously described (28). Similarly, most models failed to converge, particularly for comparisons involving ST1284 where all isolates were obtained from a single country. We then used Fisher exact tests to perform pairwise comparisons of antimicrobial and virulence genes among dominant STs. We used Mann-Whitney tests for comparison of virulence scores between dominant STs. We adjusted p values for multiple comparisons within each outcome by using the false discovery rate (29). We defined statistical significance as $p \geq 0.05$.

Results

Global Distribution of Carbapenemases

Overall, 218 isolates were positive for a single carbapenemase and 11 isolates were positive for 2 carbapenemases (Table). The OXA-48-like (n = 106) were the most common carbapenemases, followed by NDMs (n = 77), KPCs (n = 50), VIMs (n = 5), and IMPs (n = 2). The OXA-48-like carbapenemases consisted of OXA-48 (n = 41), OXA-181 (n = 55), OXA-244 (n = 3), and OXA-232 (n = 7). *E. coli* with OXA-48, OXA-181, and OXA-232 had a global distribution. OXA-244 was limited to Egypt (Table). The NDMs consisted of NDM-1 (n = 22), NDM-4 (n = 1), NDM-5 (n = 48), NDM-6 (n = 1), and NDM-7 (n = 5). *E. coli* with NDM-1 and NDM-5 had a global distribution. NDM-4 was limited to Vietnam and NDM-6 to Guatemala; NDM-7 was found in the Philippines and Vietnam (Table). The KPCs consisted of KPC-2 (n = 35), KPC-3 (n = 14), and KPC-18 (n = 1). *E. coli* with KPC-2 and KPC-3 had a global distribution, and KPC-18 was obtained from the United States (Table). *E. coli* with VIMs (VIM-1 and VIM-23) were found in Greece, Spain, Mexico, and Egypt; *E. coli* with IMP-59 were obtained from Australia (Table).

Global Distribution of Dominant *E. coli* Sequence Types and Clades

We identified 2 major STs (ST410 [20%] and ST131 [12%]) and 3 minor STs (ST1284 [8%], ST167 [7%], and ST405 [5%]) among this collection. The next most common STs did not fulfill the definition of a dominant ST: ST38 (n = 10 [4%]), ST354 (n = 7 [3%]), ST361 (n = 9 [4%]), ST648 (n = 8 [4%]), and ST11260 (n = 6 [3%]).

ST410 was the most common ST (n = 45/229 [20%]) and was positive for KPC-2 (7%), NDM-5 (27%), NDM-7 (2%), OXA-48 (4%), OXA-181 (47%), and VIM-23 [4%] (Appendix Table 1). ST410 belonged to 2 subclades: B3/H24Rx (n = 10) and B4/H24RxC (n = 35) (21).

ST131 was the second most common ST (n = 26/229 [12%]) and was positive for KPC-2 (n = 8), KPC-3 (n = 7), KPC-18 (n = 1), NDM-1 (n = 5), NDM-5 (n = 1), OXA-48 (n = 2), OXA-181 (n = 1), and OXA-232 (n = 1). One NDM-1 isolate was also positive for VIM-1. ST131 belonged to clade A/H41 (n = 2) and subclades C1_nonM27 (n = 10), C1_M27 (n = 4), and C2 (n = 10). We also note the global distribution of different minor STs (ST1284, ST167, ST405) and their clades (Appendix).

AMR Determinants and Plasmid Replicon Types

We determined quinolone resistance-determining regions mutations, β -lactamases (noncarbapenemases), aminoglycoside modifying enzymes, and plasmid replicon types among the different *E. coli* STs (Appendix Table 1). TEM-1, CTX-M-15, *aac(6')-Ib-cr*, and *sul1* were common among isolates.

Virulence Associated Factors

We assessed the presence of 37 putative virulence factors among the different dominant STs (Appendix Table 2). The following factors were present among most of isolates: *fimH* (100%), *fyuA* (55%), *traT* (64%), and *iss* (52%). Some virulence factors were associated with certain STs: *papA* (81%), *iha* (77%), *sat* (81%), *fyuA* (100%) *usp* (100%), *ompT* (100%), and *malX* (100%) with ST131, and *astA* (100%) and *iutA* (100%) with ST1284. ST131 had the highest overall number of virulence genes (n = 11), and ST410 had the lowest number of virulence genes (n = 2) (Appendix Table 2).

Carbapenemase Gene Flanking Regions and Plasmid Analysis

Because of the limitations of short-read sequencing (30), analyses of the immediate carbapenemase gene flanking regions and plasmids harboring carbapenemase genes were insufficient, especially for *bla*_{OXA-48} and *bla*_{VIMs}. We obtained results for 20/22 of *bla*_{NDM-1'}, 2/2 of *bla*_{NDM-4'}, 46/48 of *bla*_{NDM-5'}, 1/1 of *bla*_{NDM-6'}, 4/5 of *bla*_{NDM-7'}, 34/35 of *bla*_{KPC-2'}, 14/14 of *bla*_{KPC-3'}, 1/1 of *bla*_{KPC-18'}, 1/41 *bla*_{OXA-48'}, 55/55 of *bla*_{OXA-181'}, 3/3 of *bla*_{OXA-244'}, and 7/7 of *bla*_{OXA-232'}.

Among *bla*_{KPC-2'}, 15 were situated in Tn4401 elements (Tn4401a [n = 4] in ST131 and ST46, Tn4401b [n = 9] in 7 STs, and Tn4401e [n = 2] in ST131 and ST1193). Nineteen were associated with non-Tn4401 mobile elements (NTM_{KPC}) (31), including 4 ST131 and 3 ST405 strains. The *bla*_{KPC-3} genes were associated with Tn4401a (n = 9), Tn4401b (n = 3), and Tn4401d (n = 2). The *bla*_{KPC-18} was located on a novel Tn4401 variant (186 bp deletion). The *bla*_{NDM}s were located on truncated Tn125

elements, and the *bla*_{NDM} upstream regions showed substantial diversities with various IS element insertions (e.g., IS630, IS*Aba*125, IS1, and IS903 with *bla*_{NDM-1}; IS*Ecp*1 and IS1 with *bla*_{NDM-5}; and IS5 with *bla*_{NDM-7}).

All *bla*_{OXA-232} genes were located on the same 6.1 kb colKp3 plasmids (pOXA-232) (32). Sequence similarities (95%–100%) of *bla*_{KPC-3} isolates with previously sequenced plasmids in GenBank showed that most (n = 9) were harbored within IncFIB*Q*l plasmids (33); 2 KPC-3 genes were within pKPC-CAV1193 (34), 1 *bla*_{KPC-3} was within the IncFIA plasmid pBK30683 (35), and 1 *bla*_{KPC-3} was in the IncI2 plasmid pBK15692 (36). The *bla*_{OXA-181} (n = 55) were situated within Tn2013 harbored on the identical IncX3 plasmids with 99%–100% similarities to plasmid p72_X3_OXA181 (37). p72_X3_OXA181 contained the IncX3 and truncated ColKp3 replicons (13).

Discussion

A World Health Organization report showed the lack of adequate surveillance programs in many parts of the world, especially from lower- and middle-income countries (LMICs) (38). That report identified bacteria, including carbapenem-resistant *E. coli*, where global surveillance data are urgently required. LMICs bear a considerable share of the disease burden attributable to MDR *E. coli* but lack adequate genomic surveillance systems (39). Our study aimed to describe the global molecular epidemiology of 229 carbapenemase-producing *E. coli* obtained from 36 countries (including 20 LMICs) during 2015–2017. Isolates with multiple AMR genes dominated the population. The most common carbapenemase group was the OXA-48-like carbapenemases (44%), followed by NDMs (32%), KPCs (21%), VIMs (2%), and IMPs (1%). OXA-48-like carbapenemases were numerous in Egypt, Jordan, and Turkey; NDMs were numerous in Egypt, Thailand, and Vietnam, and KPCs were numerous in Colombia, Italy, and the United States.

We identified 5 dominant STs and their respective clades and subclades; 4 were global: ST410 subclades B3/H24Rx and B4/H24RxC; ST131 clade A/H41, subclades C1_nonM27/H30, C1_M27/H30, and C2/H30; ST167 subclades B1, B2, and B3; and ST405 clades A and B (Appendix Figure). ST1284 (1 clade) was limited to Turkey, and the ST167-A clade was limited to Guatemala. Dominant STs and their respective clades and subclades were associated with different underlying mobile genetic elements: ST410 was linked with NDM-5 and OXA-181; ST131 was linked with KPCs, ST1284 was linked with OXA-181, ST167 was linked with NDM-5, and ST405 was linked with various carbapenemases.

A recent survey of global carbapenemase-producing *E. coli* for the period 2002–2017 included 343 carbapenem-resistant isolates obtained mainly from the United States (40). KPC (16%), NDM (16%), and OXA-48-like (13%) carbapenemases were common. The study screened for different *E. coli* phylogroups and certain STs (ST131, ST648, and ST405). Phylogroup B2 isolates were common, and phylogroup A was dominant in Asia. Global ST131 with *bla*_{KPC}s was the most common ST, followed by ST648 with *bla*_{OXA-48-like} and ST405 with *bla*_{NDM}s.

The most frequent individual carbapenemases in our survey were OXA-181 (23%), NDM-5 (20%), OXA-48 (17%), KPC-2 (15%), and NDM-1 (10%). This result was different from carbapenemase-producing *K. pneumoniae* and *Enterobacter cloacae* complex with carbapenemases obtained from the same surveillance programs (14,41). The *K. pneumoniae* population was dominated by ST258 with KPC-2 from Greece and KPC-3 from the United States (41). The *E. cloacae* complex isolates (various STs) were dominated by VIM-1 from Greece and Italy (14). *K. pneumoniae* (42) and *E. cloacae* complex (43) are mainly hospital pathogens, whereas *E. coli* was mainly a community pathogen (6), which could partly be responsible for the different carbapenemase types among these species.

Molecular-based surveillance studies have shown that OXA-48-like enzymes are common among global carbapenemase-producing Enterobacterales (4,5). OXA-48 is currently the most common OXA-48-like derivative and OXA-181 the second most common derivative (44). OXA-48 is endemic in North Africa, Middle East, and Turkey (44). *E. coli* with *bla*_{OXA-48} is linked to various STs (44). In our study, OXA-48 was identified among 18 STs from 15 countries. *E. coli* with *bla*_{OXA-48} was common in Turkey, where it was linked with ST11260.

OXA-181 is linked with certain *E. coli* STs, especially ST410 (44). *E. coli* ST410 belongs to phylogroup A and is divided into 2 clades (A/H53 and B/H24). Clade B is divided into subclades B1/H24, B2/H24R, B3/H24Rx, and B4/H24RxC (21). The B2/H24R subclade is associated with fluoroquinolone resistance, B3/H24Rx with *bla*_{CTX-M-15}, and B4/H24RxC with *bla*_{OXA-181} (21). In our survey, OXA-181 was identified among 11 different STs obtained from 12 countries. All the OXA-181 genes were situated within Tn2013 harbored on near identical IncX3 plasmids (≈100% similarly to p72_X3_OXA181). *K. pneumoniae* ST307 with p72_X3_OXA181 was previously responsible for large outbreaks in South Africa (13,37). *E. coli* with *bla*_{OXA-181} was frequent in Jordan, Egypt (linked with ST410- B4/H24RxC subclade), and Turkey (linked with ST1284). The ST410-B4/H24RxC subclade with

*bla*_{OXA-181} was also found in Thailand and South Korea. The ST410-B3/H24Rx subclade with *bla*_{OXA-181} was present in South Africa and Kuwait.

Molecular-based surveillance studies have shown that NDMs are often the most common carbapenemase in certain regions (e.g., the Indian subcontinent) (4,5). NDM-1 is the most frequent NDM enzyme and associated with various STs within diverse plasmid platforms (45). In our survey, NDM-1 was identified among 14 different STs obtained from 10 countries. *E. coli* with *bla*_{NDM-1} was not linked with a specific ST and was evenly distributed among the different countries.

E. coli with NDM-5 is numerous among *E. coli* with NDMs from India, China, and sub-Saharan Africa (45). NDM-5, in our survey, was found among 9 different STs from 9 countries. It was common in Egypt (linked with ST410 [B4/H24Rx] and ST167 [B1 and B3]), Thailand (linked with ST410 [B4/H24Rx] and ST167 [B2]), and Vietnam (linked with ST448). ST167 belongs to phylogroup A and is an emerging carbapenemase clone associated with *bla*_{NDM-5} (46). We divided ST167 into 2 clades (A and B) and 3 subclades (B1, B2, and B3). Subclade B3 was the most dominant clade and associated with *bla*_{NDM-5} obtained from Egypt and Italy. Other subclades were less common and linked with *bla*_{NDM-5'}, *bla*_{NDM-1'}, and *bla*_{OXA-181} obtained in Guatemala (clade A), Egypt (subclades B1 and B2), and Thailand (subclade B2).

E. coli with *bla*_{KPC} is associated with ST131 (47) on diverse plasmid platforms (48). *E. coli* ST131 is global MDR high-risk clone associated with fluoroquinolone resistance and *bla*_{CTX-M}s (49). ST131 belongs to clades A/H41, B/H22, and C/H30 (50). C/H30 is divided into subclades C0, C1_nonM27, C1_M27, and C2. In our survey, KPC genes were found among 26 different STs from 11 countries. *E. coli* with *bla*_{KPC} was common in Colombia linked with various STs. ST131 was responsible for 32% of KPC isolates and obtained from Italy, Israel, Guatemala, Puerto Rico, and the United States (including Puerto Rico). ST131 with *bla*_{KPC} was dominated by the C1_nonM27 subclade. This dominance is different from that observed by Johnson et al. study (40), where the C2 subclade was common. The ST131-C1_M27 subclade in our survey was positive for *bla*_{NDM-1} and *bla*_{OXA-232}.

Among this study's strengths is that it included a large global collection of recent isolates representing multiple LMICs. We characterized all isolates using short-read WGS and provided novel information regarding the geographic distribution and MDR determinants of dominant STs and their respective clades and subclades (e.g., global ST410 was linked with

*bla*_{OXA-181}, ST131 with *bla*_{KPC}, ST167 with *bla*_{NDM-5'} and ST405 with various carbapenemases).

We showed that the underlying molecular epidemiology within the same carbapenemase groups were very different (e.g., NDM-1 was linked with various STs, including ST131-C2/H30, whereas NDM-5 was linked with ST167-B and ST410B4/H24Rx). The geographic distribution of isolates with NDM-1 and NDM-5 was different (e.g., NDM-1 showed global distribution whereas those with NDM-5 were numerous in Egypt, Thailand, and Vietnam). Similar differences were described for isolates with OXA-48 (various STs) and OXA-181 (linked with ST410B4/H24Rx). Future genomic surveys should use methodologies that characterize individual carbapenemases.

We also showed that global *bla*_{OXA-181} was harbored on near identical IncX3 plasmids (irrespective of the ST or geographic location). This finding suggests that highly similar IncX3 plasmids were mainly responsible for the global distribution of OXA-181 genes, the most common carbapenemase in this collection. The control of such IncX3 plasmids should be a public health priority.

Limitations of this study include the fact that flanking regions and plasmids harboring carbapenemases were not fully reconstructed because of the limitations of short-read sequencing (30). The characterization of plasmids is vital to fully comprehend the molecular epidemiology of global carbapenemase-producing *E. coli*, and a follow-up study using long-read sequencing is under way. Several countries included only few isolates (Table) and therefore may not be fully representative of what carbapenemase-producing *E. coli* dominates in that region.

In summary, the global carbapenemase-producing *E. coli* population is dominated by diverse STs with different characteristics and varied geographic distributions. This characterization was especially apparent within certain carbapenemases groups (i.e., NDM-1 vs. NDM-5 or OXA-48 vs. OXA-181). Ongoing genomic surveillance to characterize individual carbapenemases will assist in designing management and prevention strategies to help curtail the spread of AMR bacteria.

Acknowledgments

We thank Merck and AstraZeneca for providing the SMART and INFORM isolates.

This work was supported by a research grant from the Joint Programming Initiative on Antimicrobial Resistance-Canadian Institute Health Research program (grant no. 10016015) and the US National Institutes of Health (grant no. 10028552).

About the Author

Dr. Peirano is a research associate at Alberta Precision Laboratories and the University of Calgary. Her research interests include the molecular epidemiology of antimicrobial-resistant organisms.

References

- Tompkins K, van Duin D. Treatment for carbapenem-resistant Enterobacteriales infections: recent advances and future directions. *Eur J Clin Microbiol Infect Dis*. 2021; 40:2053–68. <https://doi.org/10.1007/s10096-021-04296-1>
- Tacconelli E, Carrara E, Savoldi A, Harbarth S, Mendelson M, Monnet DL, et al.; WHO Pathogens Priority List Working Group. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. *Lancet Infect Dis*. 2018;18:318–27. [https://doi.org/10.1016/S1473-3099\(17\)30753-3](https://doi.org/10.1016/S1473-3099(17)30753-3)
- Pitout JD, Nordmann P, Poirel L. Carbapenemase-producing *Klebsiella pneumoniae*, a key pathogen set for global nosocomial dominance. *Antimicrob Agents Chemother*. 2015;59:5873–84. <https://doi.org/10.1128/AAC.01019-15>
- Karlowsky JA, Lob SH, Kazmierczak KM, Young K, Motyl MR, Sahn DF. In vitro activity of imipenem/relebactam against *Enterobacteriaceae* and *Pseudomonas aeruginosa* isolated from intraabdominal and urinary tract infection samples: SMART Surveillance United States 2015–2017. *J Glob Antimicrob Resist*. 2020;21:223–8. <https://doi.org/10.1016/j.jgar.2019.10.028>
- Kazmierczak KM, Karlowsky JA, de Jonge BLM, Stone GG, Sahn DF. Epidemiology of carbapenem resistance determinants identified in meropenem-nonsusceptible *Enterobacteriales* collected as part of a global surveillance program, 2012 to 2017. *Antimicrob Agents Chemother*. 2021;65:e0200020. <https://doi.org/10.1128/AAC.02000-20>
- Pitout JD. Extraintestinal pathogenic *Escherichia coli*: an update on antimicrobial resistance, laboratory diagnosis and treatment. *Expert Rev Anti Infect Ther*. 2012;10:1165–76. <https://doi.org/10.1586/eri.12.110>
- Pitout JDD. Population dynamics of *Escherichia coli* causing bloodstream infections over extended time periods. *MSphere*. 2021;6:e0095621. <https://doi.org/10.1128/msphere.00956-21>
- Léger A, Lambraki I, Graells T, Cousins M, Henriksson PJG, Harbarth S, et al. Characterizing social-ecological context and success factors of antimicrobial resistance interventions across the One Health spectrum: analysis of 42 interventions targeting *E. coli*. *BMC Infect Dis*. 2021;21:873. <https://doi.org/10.1186/s12879-021-06483-z>
- Nordmann P, Poirel L. The difficult-to-control spread of carbapenemase producers among *Enterobacteriaceae* worldwide. *Clin Microbiol Infect*. 2014;20:821–30. <https://doi.org/10.1111/1469-0691.12719>
- Manges AR, Geum HM, Guo A, Edens TJ, Fibke CD, Pitout JDD. Global extraintestinal pathogenic *Escherichia coli* (ExPEC) lineages. *Clin Microbiol Rev*. 2019;32:e00135–18. <https://doi.org/10.1128/CMR.00135-18>
- Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing. Twenty-fifth information supplement (M100–S25). Wayne (PA): The Institute; 2015.
- Holland MS, Nobrega D, Peirano G, Naugler C, Church DL, Pitout JDD. Molecular epidemiology of *Escherichia coli* causing bloodstream infections in a centralized Canadian region: a population-based surveillance study. *Clin Microbiol Infect*. 2020;26:1554.e1–e8.
- Lowe M, Kock MM, Coetzee J, Hoosien E, Peirano G, Strydom KA, et al. *Klebsiella pneumoniae* ST307 with *bla*_{OXA-181} South Africa, 2014–2016. *Emerg Infect Dis*. 2019;25:739–47. <https://doi.org/10.3201/eid2504.181482>
- Peirano G, Matsumura Y, Adams MD, Bradford P, Motyl M, Chen L, et al. Genomic epidemiology of global carbapenemase-producing *Enterobacter* spp., 2008–2014. *Emerg Infect Dis*. 2018;24:1010–9. <https://doi.org/10.3201/eid2406.171648>
- Nurk S, Bankevich A, Antipov D, Gurevich AA, Korobeynikov A, Lapidus A, et al. Assembling single-cell genomes and mini-metagenomes from chimeric MDA products. *J Comput Biol*. 2013;20:714–37. <https://doi.org/10.1089/cmb.2013.0084>
- Zankari E, Hasman H, Cosentino S, Vestergaard M, Rasmussen S, Lund O, et al. Identification of acquired antimicrobial resistance genes. *J Antimicrob Chemother*. 2012;67:2640–4. <https://doi.org/10.1093/jac/dks261>
- Carattoli A, Zankari E, García-Fernández A, Voldby Larsen M, Lund O, Villa L, et al. In silico detection and typing of plasmids using PlasmidFinder and plasmid multilocus sequence typing. *Antimicrob Agents Chemother*. 2014;58:3895–903. <https://doi.org/10.1128/AAC.02412-14>
- Zhou Z, Alikhan NF, Mohamed K, Fan Y, Achtman M; Agama Study Group. The Enterobase user's guide, with case studies on *Salmonella* transmissions, *Yersinia pestis* phylogeny, and *Escherichia coli* core genomic diversity. *Genome Res*. 2020;30:138–52. <https://doi.org/10.1101/gr.251678.119>
- Joensen KG, Scheutz F, Lund O, Hasman H, Kaas RS, Nielsen EM, et al. Real-time whole-genome sequencing for routine typing, surveillance, and outbreak detection of verotoxigenic *Escherichia coli*. *J Clin Microbiol*. 2014;52:1501–10. <https://doi.org/10.1128/JCM.03617-13>
- Matsumura Y, Pitout JDD, Peirano G, DeVinney R, Noguchi T, Yamamoto M, et al. Rapid identification of different *Escherichia coli* sequence type 131 clades. *Antimicrob Agents Chemother*. 2017;61:e00179-17. <https://doi.org/10.1128/AAC.00179-17>
- Roer L, Overballe-Petersen S, Hansen F, Schønning K, Wang M, Røder BL, et al. *Escherichia coli* sequence type 410 is causing new international high-risk clones. *MSphere*. 2018;3:e00337-18. <https://doi.org/10.1128/msphere.00337-18>
- Wang M, Earley M, Chen L, Hanson BM, Yu Y, Liu Z, et al. Clinical outcomes and bacterial characteristics of carbapenem-resistant *Klebsiella pneumoniae* complex among patients from different global regions (CRACKLE-2): a prospective, multicentre, cohort study. *Lancet Infect Dis*. 2021 Nov 9 [Epub ahead of print]. [https://doi.org/10.1016/S1473-3099\(21\)00399-6](https://doi.org/10.1016/S1473-3099(21)00399-6)
- Stamatakis A. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*. 2014;30:1312–3. <https://doi.org/10.1093/bioinformatics/btu033>
- Tonkin-Hill G, Lees JA, Bentley SD, Frost SDW, Corander J. RhierBAPS: An R implementation of the population clustering algorithm hierBAPS. *Wellcome Open Res*. 2018; 3:93. <https://doi.org/10.12688/wellcomeopenres.14694.1>
- Cheng L, Connor TR, Sirén J, Aanensen DM, Corander J. Hierarchical and spatially explicit clustering of DNA sequences with BAPS software. *Mol Biol Evol*. 2013;30:1224–8. <https://doi.org/10.1093/molbev/mst028>
- Letunic I, Bork P. Interactive tree of life (iTOL) v3: an online tool for the display and annotation of phylogenetic and other

- trees. *Nucleic Acids Res.* 2016;44(W1):W242-5. <https://doi.org/10.1093/nar/gkw290>
27. R Core Team. R: a language and environment for statistical computing. Vienna: R Foundation for Statistical Computing; 2017.
 28. Troxler S, Lalonde T, Wilson JR. Exact logistic models for nested binary data. *Stat Med.* 2011;30:866–76. <https://doi.org/10.1002/sim.4157>
 29. Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J R Stat Soc B.* 1995;57:289–300. <https://doi.org/10.1111/j.2517-6161.1995.tb02031.x>
 30. Arredondo-Alonso S, Willems RJ, van Schaik W, Schürch AC. On the (im)possibility of reconstructing plasmids from whole-genome short-read sequencing data. *Microb Genom.* 2017;3:e000128. <https://doi.org/10.1099/mgen.0.000128>
 31. Chen L, Mathema B, Chavda KD, DeLeo FR, Bonomo RA, Kreiswirth BN. Carbapenemase-producing *Klebsiella pneumoniae*: molecular and genetic decoding. *Trends Microbiol.* 2014;22:686–96. <https://doi.org/10.1016/j.tim.2014.09.003>
 32. Potron A, Rondinaud E, Poirel L, Belmonte O, Boyer S, Camiade S, et al. Genetic and biochemical characterisation of OXA-232, a carbapenem-hydrolysing class D β -lactamase from *Enterobacteriaceae*. *Int J Antimicrob Agents.* 2013;41:325–9. <https://doi.org/10.1016/j.ijantimicag.2012.11.007>
 33. García-Fernández A, Villa L, Carta C, Venditti C, Giordano A, Venditti M, et al. *Klebsiella pneumoniae* ST258 producing KPC-3 identified in Italy carries novel plasmids and OmpK36/OmpK35 porin variants. *Antimicrob Agents Chemother.* 2012;56:2143–5. <https://doi.org/10.1128/AAC.05308-11>
 34. Sheppard AE, Stoesser N, Sebra R, Kasarskis A, Deikus G, Anson L, et al. Complete genome sequence of KPC-producing *Klebsiella pneumoniae* strain CAV1193. *Genome Announc.* 2016;4:e01649-15. <https://doi.org/10.1128/genomeA.01649-15>
 35. Chen L, Chavda KD, Melano RG, Hong T, Rojzman AD, Jacobs MR, et al. Molecular survey of the dissemination of two *bla*_{KPC}-harboring IncFIA plasmids in New Jersey and New York hospitals. *Antimicrob Agents Chemother.* 2014;58:2289–94. <https://doi.org/10.1128/AAC.02749-13>
 36. Chen L, Chavda KD, Al Laham N, Melano RG, Jacobs MR, Bonomo RA, et al. Complete nucleotide sequence of a *bla*_{KPC}-harboring IncI2 plasmid and its dissemination in New Jersey and New York hospitals. *Antimicrob Agents Chemother.* 2013;57:5019–25. <https://doi.org/10.1128/AAC.01397-13>
 37. Strydom KA, Chen L, Kock MM, Stoltz AC, Peirano G, Norega DB, et al. *Klebsiella pneumoniae* ST307 with OXA-181: threat of a high-risk clone and promiscuous plasmid in a resource-constrained healthcare setting. *J Antimicrob Chemother.* 2020;75:896–902. <https://doi.org/10.1093/jac/dkz550>
 38. World Health Organization. Antimicrobial resistance: global report on surveillance 2014. Geneva: World Health Organization; 2014. p. 257.
 39. Acharya KP, Subramanya SH, Pitout JDD. Inclusion of next-generation leaders and cost-effective precision diagnostic techniques are vital in combatting antimicrobial resistance in low- and middle-income countries. *JAC Antimicrob Resist.* 2020 Jun 23 [Epub ahead of print]. <https://doi.org/10.1093/jacamr/dlaa032>
 40. Johnston BD, Thuras P, Porter SB, Anacker M, VonBank B, Vagnone PS, et al. Global molecular epidemiology of carbapenem-resistant *Escherichia coli* (2002–2017). *Eur J Clin Microbiol Infect Dis.* 2021 Jul 19 [Epub ahead of print]. <https://doi.org/10.1007/s10096-021-04310-6>
 41. Peirano G, Bradford PA, Kazmierczak KM, Chen L, Kreiswirth BN, Pitout JD. Importance of clonal complex 258 and IncF_{K2-like} plasmids among a global collection of *Klebsiella pneumoniae* with *bla*_{KPC}. *Antimicrob Agents Chemother.* 2017;61:e02610-16. <https://doi.org/10.1128/AAC.02610-16>
 42. Peirano G, Chen L, Kreiswirth BN, Pitout JDD. Emerging antimicrobial-resistant high-risk *Klebsiella pneumoniae* clones ST307 and ST147. *Antimicrob Agents Chemother.* 2020;64:e01148-20. <https://doi.org/10.1128/AAC.01148-20>
 43. Stokes W, Peirano G, Matsumura Y, Nobrega D, Pitout JDD. Population-based surveillance of *Enterobacter cloacae* complex causing blood stream infections in a centralized Canadian region. *Eur J Clin Microbiol Infect Dis.* 2021 Jul 14 [Epub ahead of print]. <https://doi.org/10.1007/s10096-021-04309-z>
 44. Pitout JDD, Peirano G, Kock MM, Strydom KA, Matsumura Y. The global ascendancy of OXA-48-type carbapenemases. *Clin Microbiol Rev.* 2019;33:e00102-19. <https://doi.org/10.1128/CMR.00102-19>
 45. Wu W, Feng Y, Tang G, Qiao F, McNally A, Zong Z. NDM metallo- β -lactamases and their bacterial producers in health care settings. *Clin Microbiol Rev.* 2019;32:e00115-18. <https://doi.org/10.1128/CMR.00115-18>
 46. Cummins EA, Snaith AE, McNally A, Hall RJ. The role of potentiating mutations in the evolution of pandemic *Escherichia coli* clones. *Eur J Clin Microbiol Infect Dis.* 2021 Nov 17 [Epub ahead of print]. <https://doi.org/10.1007/s10096-021-04359-3>
 47. Peirano G, Bradford PA, Kazmierczak KM, Badal RE, Hackel M, Hoban DJ, et al. Global incidence of carbapenemase-producing *Escherichia coli* ST131. *Emerg Infect Dis.* 2014;20:1928–31. <https://doi.org/10.3201/eid2011.141388>
 48. Stoesser N, Sheppard AE, Peirano G, Anson LW, Pankhurst L, Sebra R, et al. Genomic epidemiology of global *Klebsiella pneumoniae* carbapenemase (KPC)-producing *Escherichia coli*. *Sci Rep.* 2017;7:5917. <https://doi.org/10.1038/s41598-017-06256-2>
 49. Mathers AJ, Peirano G, Pitout JD. The role of epidemic resistance plasmids and international high-risk clones in the spread of multidrug-resistant *Enterobacteriaceae*. *Clin Microbiol Rev.* 2015;28:565–91. <https://doi.org/10.1128/CMR.00116-14>
 50. Pitout JDD, Finn TJ. The evolutionary puzzle of *Escherichia coli* ST131. *Infect Genet Evol.* 2020;81:104265. <https://doi.org/10.1016/j.meegid.2020.104265>

Address for correspondence: Johann D.D. Pitout, University of Calgary, #9, 3535 Research Rd NW, Calgary, AB T2L 2K8, Canada; email: jpitout@ucalgary.ca

Genomic Epidemiology of Global Carbapenemase-Producing *Escherichia coli*, 2015–2017

Appendix

Details of the SMART and INFORM surveillance programs

The Merck Study for Monitoring Antimicrobial Resistance Trends (SMART) program (2015–17) included isolates from intra-abdominal and urinary tract infections from the following 55 countries: Egypt, Morocco, Kenya, South Africa, Tunisia (Africa); China, Hong Kong, India, Japan, Malaysia, Singapore, South Korea, Taiwan, Thailand, Vietnam (Asia); Croatia, Czech Republic, Estonia, France, Georgia, Greece, Germany, Hungary, Italy, Latvia, Lithuania, Portugal, Romania, Serbia, Slovenia, Spain, Switzerland, Turkey, United Kingdom (Europe); Argentina, Brazil, Chile, Colombia, Dominican Republic, Ecuador, Guatemala, Mexico, Puerto Rico, Panama, Uruguay, Venezuela (Latin America); Jordan, Lebanon, Israel, Saudi Arabia, UAE (Middle East); Canada, United States (North America); and Australia, New Zealand, Philippines (South Pacific).

The INFORM surveillance program of antimicrobial resistance (2015–17) included isolates from intra-abdominal, urinary tract, blood, skin and soft tissue and lower respiratory tract infections from the following 42 countries: Egypt, Kenya, Nigeria, South Africa (Africa); China, South Korea, Taiwan, Thailand (Asia); Austria, Belgium, Bulgaria, Greece, Czech Republic, Denmark, France, Germany, Hungary, Italy, Macedonia, Portugal, Poland, Russia, Romania, Slovakia, Spain, Turkey, United Kingdom (Europe); Argentina, Brazil, Chile, Colombia, Mexico, Uruguay, Venezuela (Latin America); Lebanon, Israel, Syria, Kuwait (Middle East); United States (North America); and Australia, Philippines, Japan (South Pacific).

Both programs collected 100 consecutive clinically relevant non-repeat Gram-negative bacteria per annum from each institution.

Global distribution of dominant *E. coli* Sequence types (STs) and clades

ST410

The ST410-B3/H24Rx subclade showed a global distribution (Georgia [n = 1] with OXA-48, Kuwait [n = 1] with OXA-181, Mexico [n = 2] with VIM-23, Morocco [n = 1] with OXA-48, South Africa [n = 2] with OXA-181, USA [n = 2] with KPC-2 and Vietnam [n = 1] with KPC-2). The ST410-B4/H24RxC subclade was found in Jordan (n = 15 with OXA-181), Egypt (n = 11 with NDM-5 and OXA-181), Philippines (n = 1 with NDM-7), South Korea (n = 1 with NDM-5 and OXA-181), Thailand (n = 6 with NDM-5 and OXA-181 and Vietnam (n = 1 with NDM-5).

ST131

The ST131-A/H41 clade (n = 2) was positive for KPC-3 (from USA) and OXA-48 (from Lebanon). The ST131-C1_M27/H30 subclade (n = 4) was positive for NDM-1 (from Russia (n = 2), Philippines [n = 1]) and OXA-232 from Thailand (n = 1). The ST131-C1_nonM27/H30 subclade (n = 10) was positive for KPC-2 from Guatemala (n = 4), Israel (n = 2) and USA (n = 1); KPC-3 from Italy (n = 2) and KPC-18 from USA (n = 1). The ST131-C2/H30 subclade (n = 10) was positive for KPC-2 (n = 1 from Puerto Rico), KPC-3 (n = 4 from Italy), NDM-1 (n = 2 from Egypt), NDM-5 (n = 1 from Canada), OXA-48 (n = 1 from Egypt), and OXA-181 (n = 1 from Egypt). One ST131-C2 with NDM-1 was also positive for VIM-1.

ST167

The ST167-A clade (n = 2) was found in Guatemala with NDM-1, ST167-B1 subclade (n = 3) was found in Egypt with NDM-5; ST167-B2 subclade (n = 5) was found in Egypt (n = 2 with OXA-181), and Thailand (n = 3 with NDM-5); ST167-B3 subclade (n = 7) was present in Egypt (with NDM-1 [n = 1], NDM-5 [4]) and Italy (n = 2 with NDM-5).

ST405

The ST405-A clade was found in Egypt with NDM-5 and OXA-48. The ST405-B clade was present in Colombia (n = 1 with KPC-2), Spain (n = 1 with OXA-48), Vietnam (n = 4 with NDM-5 and KPC-2), Turkey (n = 2 with OXA-48) and UK (n = 1 with NDM-5).

The non-dominant STs (n = 112/229 [49%]) were positive for KPC-2, KPC-3, NDM-1, NDM-5, NDM-6, NDM-7, OXA-48, OXA-181, OXA-232, OXA-244, VIM-1 and IMP-59

(Appendix Table 1). Non-dominant STs showed a global distribution. They were found in Europe (Austria, Belgium, Georgia, Germany, Greece, Italy, Romania, Russia, Serbia, Spain, UK, Turkey), Africa (Egypt, Morocco, Tunisia), Middle East (Lebanon, Israel, Jordan, Kuwait, Saudi Arabia), Asia (Malaysia, Thailand, Philippines, Taiwan, Vietnam), Latin America (Argentina, Brazil, Colombia, Guatemala, Mexico, Puerto Rico, Venezuela), North America (USA) and Australia. Non-dominant STs were common in Turkey where they were linked with OXA-48; in Egypt where they were linked with NDM-1, NDM-5, OXA-181, OXA-244, and OXA-48; and Vietnam where they were linked with KPC-2, NDM-1, NDM-5, NDM-7, OXA-48 and OXA-181.

Appendix Table 1. Characteristics of dominant Sequence Types among carbapenemase-producing *Escherichia coli*

Characteristic	ST410 n = 45 (%)	ST131 n = 26 (%)	ST1284 n = 18 (%)	ST167 n = 17 (%)	ST405 n = 11 (%)	Other STs [#] n = 112 (%)	All STs n = 229 (%)
Geographic location	Global	Global	Turkey	Global	Global	Global	Global
QRDR mutations							
<i>gyrA</i> S83L	44 (98)	26 (100)	18 (100)	17 (100)	10 (91)	76 (68)	191 (83)
<i>gyrA</i> D87N	44 (98)	25 (96)	18 (100)	17 (100)	10 (91)	61 (54)	175 (77)
<i>parC</i> S80I	44 (98)	25 (96)	18 (100)	17 (100)	11 (100)	62 (55)	177 (78)
<i>parC</i> E84V	0 ^a	24 (92) ^b	0 ^a	0 ^a	0 ^a	1 (1)	25 (12)
<i>parE</i> I529L	0 ^a	25 (96) ^b	0 ^a	0 ^a	0 ^a	1 (1)	26 (11)
Carbapenemases							
KPC-2	3 (7) ^a	8 (31) ^b	0 ^a	0 ^a	3 (27)	21 (19)	35 (15)
KPC-3	0 ^a	7 (27) ^b	0 ^a	0 ^a	0	7 (6)	14 (6)
KPC-18	0	1 (4)	0	0	0	0	1 (0.4)
NDM-1	0 ^a	4 (15) ^b	0	3 (18)	0	12 (11)	19 (8)
NDM-4	0	0	0	0	1 (9)	0	1 (0.4)
NDM-5	12 (27) ^a	1 (4) ^b	0 ^b	11 (65) ^c	3 (27)	13 (12)	40 (17)
NDM-6	0	0	0	0	0	1 (1)	1 (0.4)
NDM-7	1 (2)	0	0	0	0	4 (4)	5 (2)
OXA-48	2 (4)	2 (8)	0	0	4 (36)	32 (29)	40 (17)
OXA-181	21 (47) ^a	1 (4) ^b	18 (100) ^c	2 (12) ^b	0 ^b	6 (5)	48 (21)
OXA-232	0	1 (4)	0	0	0	4 (4)	5 (2)
OXA-244	0	0	0	0	0	3 (3)	3 (1)
VIM-1	0	0	0	0	0	2 (2)	2 (1)
VIM-23	2 (4)	0	0	0	0	0	2 (1)
IMP-59	0	0	0	0	0	2 (2)	2 (1)
NDM-1 + VIM-1	0	1 (4)	0	0	0	0	1 (0.4)
NDM-1 + OXA-181	0	0	0	0	0	2 (2)	2 (1)
NDM-5 + OXA-48	0	0	0	1 (6)	0	0	1 (0.4)
NDM-5 + OXA-181	4 (9)	0	0	0	0	1 (1)	5 (2)
NDM-5 + OXA-232	0	0	0	0	0	2 (2)	2 (1)
Other β-lactamases*							
OXAs	21 (47) ^a	11 (42) ^a	17 (94) ^b	7 (41) ^a	3 (27) ^a	23 (21)	82 (36)
CMYs	40 (89) ^a	2 (8) ^b	18 (100) ^a	0 ^b	2 (18) ^b	18 (16)	80 (35)
DHA-1	0	0	0	0	0	3 (3)	3 (1)
CTX-Ms	38 (84) ^a	13 (50) ^b	11 (61)	13 (77)	9 (82)	58 (52)	142 (62)

Characteristic	ST410 n = 45 (%)	ST131 n = 26 (%)	ST1284 n = 18 (%)	ST167 n = 17 (%)	ST405 n = 11 (%)	Other STs [#] n = 112 (%)	All STs n = 229 (%)
TEM-1	35 (78) ^a	18 (69) ^{a,c}	0 ^b	8 (47) ^{c,d}	3 (27) ^{b,d}	79 (71)	143 (62)
TEMs (ESBLs)	1 (2)	0	0	1 (6)	0	2 (2)	4 (2)
SHV-2	0	0	0	0	0	1 (1)	1 (0.4)
SHV-12	0	2 (8)	0	2 (12)	0	8 (7)	12 (5)
Aminoglycoside modifying enzymes							
<i>aadA1</i>	0 ^a	2 (8)	0	1 (6)	3 (27) ^b	23 (21)	29 (13)
<i>aadA2</i>	15 (33) ^a	1 (4) ^b	0 ^b	8 (47) ^a	3 (27)	13 (12)	40 (17)
<i>aadA2b</i>	0	0	0	0	0	6 (5)	6 (3)
<i>aadA22</i>	0 ^a	0 ^a	0 ^a	5 (29) ^b	0	0	5 (2)
<i>aadA5</i>	27 (60) ^a	11 (42) ^a	18 (100) ^b	7 (41) ^a	5 (46) ^a	34 (30)	102 (45)
<i>aadA16</i>	0	0	0	0	1 (9)	4 (4)	5 (2)
<i>aac(3')-IIa</i>	2 (4) ^a	2 (8) ^a	18 (100) ^b	1 (6) ^a	2 (18) ^a	18 (16)	43 (19)
<i>acc(3')-IIId</i>	31 (69) ^a	3 (12) ^b	0 ^b	2 (12) ^b	1 (9) ^b	20 (18)	57 (25)
<i>aac(6')-Ib</i>	0	0	0	0	0	4 (4)	4 (2)
<i>aac(6')-Ib-cr</i>	41 (91) ^a	6 (23) ^b	17 (94) ^{a,c}	11 (65) ^{c,d}	5 (46) ^{b,d}	41 (37)	121 (53)
<i>aac(6')-Ia</i>	0	0	0	0	0	1 (1)	1 (0.4)
<i>aac(6')-IIa</i>	0	0	0	0	0	1 (1)	1 (0.4)
<i>aac(6')-33</i>	0	1 (4)	0	0	0	1 (1)	2 (1)
<i>ant(2')-1a</i>	0	1 (4)	0	0	0	5 (5)	6 (3)
<i>aph(3')-Ia</i>	0	1 (4)	0	0	1 (9)	6 (5)	8 (3)
<i>aph(3')-Ib</i>	15 (33) ^a	5 (19) ^a	17 (94) ^b	4 (24) ^a	1 (9) ^a	16 (14)	58 (25)
<i>aph(3')-VI</i>	0	2 (8)	0	1 (6)	0	7 (6)	10 (4)
<i>aph(4')-1a</i>	0	0	0	3 (18)	0	2 (2)	5 (2)
<i>aph(6')-1d</i>	34 (76) ^a	7 (27) ^b	14 (78) ^a	5 (29) ^b	6 (55)	38 (34)	104 (45)
<i>rmtB</i>	0 ^a	0 ^a	0 ^a	7 (41) ^b	1 (9)	7 (6)	15 (7)
<i>rmtC</i>	0	2 (8)	0	0	0	3 (3)	5 (2)
Other AMR determinants							
<i>fosA</i>	0	0	0	3 (18)	0	4 (4)	7 (3)
<i>qnrA1</i>	0	1 (4)	0	0	0	2 (2)	3 (1)
<i>qnrB1</i>	0	0	0	0	0	4 (4)	4 (2)
<i>qnrB2</i>	0	0	0	0	0	1 (1)	1 (0.4)
<i>qnrB19</i>	0	0	0	0	0	5 (5)	5 (2)
<i>qnrB4</i>	0	0	0	0	0	3 (3)	3 (1)
<i>qnrB6</i>	0	0	0	0	0	3 (3)	3 (1)
<i>qnrB9</i>	0	0	0	0	0	2 (2)	2 (1)
<i>qnrS1</i>	26 (58) ^a	4 (15) ^b	18 (100) ^c	3 (18) ^b	1 (9) ^b	18 (16)	70 (31)
<i>dfrA1</i>	0	1 (4)	0	0	0	5 (5)	6 (3)
<i>dfrA12</i>	16 (36) ^a	1 (4) ^b	0 ^b	8 (47) ^a	3 (27)	17 (15)	45 (20)
<i>dfrA14</i>	0	0	0	0	1 (9)	18 (16)	19 (8)
<i>dfrA15</i>	0	0	0	0	0	1 (1)	1 (0.4)
<i>dfrA17</i>	35 (78) ^{a,c}	11 (42) ^b	18 (100) ^a	8 (47) ^{b,c}	8 (73)	33 (30)	113 (49)
<i>dfrA27</i>	0 ^a	1 (4)	0	0	3 (27) ^b	6 (5)	10 (4)
<i>dfrA5</i>	0	0	0	0	0	3 (3)	3 (1)
<i>dfrA7</i>	0	1 (4)	0	0	0	0	1 (0.4)
<i>dfrA8</i>	0	0	0	0	0	1 (1)	1 (0.4)
<i>sul1</i>	41 (91) ^a	15 (58) ^b	18 (100) ^a	15 (88)	8 (73)	63 (56)	160 (70)
<i>sul2</i>	34 (76) ^a	8 (31) ^b	18 (100) ^c	8 (47) ^{a,b}	5 (46) ^{a,b}	40 (36)	113 (49)
<i>sul3</i>	0	0	0	1 (6)	0	13 (12)	14 (6)
<i>tetA</i>	9 (20) ^{a,b}	9 (35) ^{a,c}	0 ^b	12 (71) ^c	5 (46) ^{a,c}	27 (24)	62 (27)
<i>tetB</i>	35 (78) ^{a,c}	2 (8) ^b	4 (22) ^{b,c}	1 (6) ^b	5 (46) ^c	35 (32)	82 (36)
<i>tetD</i>	0	1 (4)	0	0	0	2 (2)	3 (1)
Plasmid types							
Col-like	44 (98) ^a	12 (46) ^b	18 (100) ^a	6 (35) ^b	5 (46) ^b	47 (42)	132 (58)
IncFIA	44 (98) ^a	16 (62) ^b	18 (100) ^a	17 (100) ^a	9 (82)	39 (35)	143 (62)
IncFIB	45 (100) ^a	24 (92) ^a	18 (100) ^a	4 (24) ^b	10 (91) ^a	83 (74)	184 (80)
IncFIC	0	0	0	0	0	10 (9)	10 (4)
IncFII	45 (100) ^a	25 (96) ^{a,c}	7 (39) ^b	14 (82) ^c	10 (91) ^{a,c}	71 (63)	172 (75)
IncI1	6 (13) ^a	3 (12) ^a	18 (100) ^b	10 (59) ^c	6 (55) ^c	30 (27)	73 (32)
IncH12	0	0	0	0	0	8 (7)	8 (3)
IncA	0	0	0	0	0	2 (2)	2 (1)
IncB/O/K/Z	0	1 (4)	0	0	0	8 (7)	9 (4)
IncC	2 (4)	4 (15)	0	0	0	13 (12)	19 (8)
IncL	2 (4) ^a	0 ^a	0 ^a	0 ^a	4 (36) ^b	20 (18)	26 (11)
IncM	0	0	0	1 (6)	0	4 (4)	5 (2)
IncN	4 (9)	2 (8)	0	0	4 (36)	14 (13)	24 (10)
IncP	0	4 (15)	0	0	0	1 (1)	5 (2)
IncQ	32 (71) ^a	0 ^b	0 ^b	0 ^b	1 (9) ^b	15 (13)	48 (21)
IncR	2 (4)	2 (8)	0	3 (18)	1 (9)	8 (7)	16 (7)

Characteristic	ST410 n = 45 (%)	ST131 n = 26 (%)	ST1284 n = 18 (%)	ST167 n = 17 (%)	ST405 n = 11 (%)	Other STs [#] n = 112 (%)	All STs n = 229 (%)
IncX1	1 (2)	1 (4)	0	0	1 (9)	5 (5)	8 (3)
IncX3	27 (60) ^a	2 (8) ^b	18 (100) ^c	10 (59) ^a	1 (9) ^b	23 (21)	81 (35)
IncX4	2 (4)	2 (8)	0	3 (18)	0	5 (5)	12 (5)
IncX5	0	0	0	0	0	1 (1)	1 (0.4)
IncU	0	0	0	0	0	2 (2)	2 (1)
IncY	9 (20)	0	0	0	0	12 (11)	21 (9)

QRDRs; quinolone resistance-determining regions;

^{a-d}Rates followed by different letters indicate significant differences between dominant STs at the 5% level (adjusted for multiple comparisons for each outcome);

[#]Other STs (no) include the following: ST10 (5), ST12 (2), ST34 (1), ST38 (10), ST44 (1), ST46 (5), ST58 (2), ST69 (3), ST73 (1), ST88 (1), ST95 (4), ST127 (1), ST141 (1), ST156 (3), ST191 (1), ST205 (1), ST244 (1), ST345 (1), ST349 (1), ST354 (7), ST357 (2), ST361 (9), ST404 (1), ST448 (4), ST538 (1), ST540 (1), ST607 (1), ST617 (4), ST624 (1), ST635 (1), ST648 (8), ST973 (1), ST1148 (1), ST1193 (2), ST1196 (1), ST1431 (1), ST1434 (1), ST1470 (1), ST1487 (1), ST1722 (1), ST2003 (2), ST2083 (2), ST2172 (1), ST2279 (1), ST3580 (1), ST4553 (1), ST5229 (1), ST6802 (1), ST11260 (6);

*Other β-lactamases (no) include the following: OXA-1 (66), OXA-9 (11), OXA-10 (5), CMY-2 (40), CMY-4 (4), CMY-6 (4), CMY-16 (1), CMY-42 (30), CMY-146 (1), CTX-M-3 (2), CTX-M-8 (1), CTX-M-9 (1), CTX-M-14 (8), CTX-M-15 (103), CTX-M-24 (12), CTX-M-27 (6), CTX-M-55 (8), CTX-M-196 (1), TEM-33 (1), TEM-35 (1), TEM-52 (1), TEM-143 (1).

Appendix Table 2. Virulence profiles associated with dominant Sequence Types among carbapenemase-producing *Escherichia coli*

Characteristic	ST410 n = 45 (%)	ST131 n = 26 (%)	ST1284 n = 18 (%)	ST167 n = 17 (%)	ST405 n = 11 (%)	Other STs [#] n = 112 (%)	All STs n = 229 (%)
Adhesions							
<i>papA</i>	0 ^a	21 (81) ^b	0 ^a	0 ^a	4 (36) ^c	24 (21)	49 (21)
<i>papC</i>	1 (2) ^a	10 (38) ^b	0 ^a	0 ^a	2 (18)	20 (18)	33 (14)
<i>iha</i>	0 ^a	20 (77) ^b	0 ^a	0 ^a	0 ^a	18 (16)	38 (17)
<i>fimH</i>	45 (100)	26 (100)	18 (100)	17 (100)	11 (100)	112 (100)	229 (100)
<i>tsh</i>	0	0	0	0	0	4 (4)	4 (2)
<i>hra</i>	11 (24) ^a	14 (54) ^b	0 ^c	15 (88) ^d	9 (82) ^{b,d}	40 (36)	89 (39)
<i>afaABCD</i>	0 ^a	5 (19) ^b	0	0	0	15 (13)	20 (9)
<i>draABCDP</i>	0	0	0	0	0	0	0
Toxins							
<i>hlyA</i>	1 (2)	0	0	0	0	23 (21)	24 (10)
<i>sat</i>	0 ^a	21 (81) ^b	0 ^a	0 ^a	0 ^a	17 (15)	38 (17)
<i>vat</i>	0	0	0	0	0	15 (13)	15 (7)
<i>astA</i>	0 ^a	2 (8) ^a	18 (100) ^b	0 ^a	0 ^a	13 (12)	33 (14)
<i>cnf1</i>	0 ^a	7 (27) ^b	0 ^a	0 ^a	0	5 (4)	12 (5)
Siderophores							
<i>iroN</i>	1 (2)	0	0	0	0	27 (24)	28 (12)
<i>fyuA</i>	15 (33) ^a	26 (100) ^b	0 ^c	9 (53) ^a	11 (100) ^b	64 (57)	125 (55)
<i>ireA</i>	0	1 (4)	0	0	0	9 (8)	10 (4)
<i>iutA</i>	7 (16) ^a	20 (77) ^b	18 (100) ^b	4 (24) ^a	3 (27) ^a	48 (43)	100 (44)
Capsular antigen							
<i>kpsM</i> II	0 ^a	18 (69) ^b	0 ^a	0 ^a	7 (64) ^b	36 (32)	61 (27)
<i>kpsM</i> III	0 ^a	0 ^a	0 ^a	0 ^a	4 (36) ^b	1 (1)	5 (2)
Miscellaneous							
<i>usp</i>	0 ^a	26 (100) ^b	0 ^a	0 ^a	0 ^a	23 (21)	49 (21)
<i>traT</i>	8 (18) ^a	24 (92) ^b	18 (100) ^b	15 (88) ^b	10 (91) ^b	71 (63)	146 (64)
<i>ompT</i>	1 (2) ^a	26 (100) ^b	0 ^a	2 (12) ^a	0 ^a	48 (43)	77 (34)
<i>iss</i>	3 (7) ^a	25 (96) ^b	18 (100) ^b	17 (100) ^b	0 ^a	56 (50)	119 (52)
<i>malX</i>	0 ^a	26 (100) ^b	0 ^a	0 ^a	0 ^a	16 (14)	42 (18)
<i>cdtB</i>	0	0	0	0	0	0	0
<i>cvaC</i>	1 (2)	0	0	0	1 (9)	20 (18)	22 (10)
<i>ibeABC</i>	0	0	0	0	0	11 (10)	11 (5)
Virulence scores*	2 (1–7) ^a	11 (8–15) ^b	5 (5–5) ^c	5 (4–6) ^c	5 (3–7) ^c	6 (1–16)	5 (1–16)

papAC, genes of P fimbriae operon; *iha*, iron-regulated adhesin; *fimH*, type-1 fimbriae; *tsh*, temperature sensitive hemagglutinin; *hra*, heat-resistant agglutinin, *afa*, afimbrial adhesin; *dra*, Dr binding adhesins; *hlyA*, α-hemolysin; *sat*, secreted autotransporter toxin; *vat*, vacuolating autotransporter toxin; *astA*, enteroaggregative *E. coli* toxin; *cnf1*, cytotoxic necrotizing factor; *iroN*, salmochelin (siderophore) receptor; *fyuA*, yersiniabactin (siderophore) receptor; *ireA*, iron-regulated element (catecholate siderophore); *iutA*, aerobactin (siderophore) receptor; *kpsM* II, group II capsule variants synthesis; *kpsM* III, group III capsule variants synthesis; *usp*, uropathogenic-specific protein; *traT*, complement inhibition protein; *ompT*, outer membrane protein T; *iss*, increased serum survival; *malX*, pathogenicity island marker; *cdtB*, cytolethal distending toxin B; *cvaC*, factor facilitating colonization; *ibeABC*, invasion of brain endothelium;

^{a-d}Rates followed by different letters indicate significant differences between dominant STs at the 5% level (adjusted for multiple comparisons for each outcome);

*The virulence gene score was the number of virulence operons detected (median; range).

