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Carbapenemase-producing *Escherichia coli* is becoming more prevalent in Spain mainly because of the polyclonal dissemination of OXA-48

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Objectives: The objective of this study was to analyse the microbiological traits and the population structure of carbapenemase-producing (CP) *Escherichia coli* isolates collected in Spain between 2012 and 2014.

Methods: Two-hundred-and-thirty-nine *E. coli* isolates non-susceptible to carbapenems were studied. The carbapenemase genes and the phylogenetic groups were characterized using PCR. MLST was carried out using the typing schemes of the University of Warwick and the Institut Pasteur. The diversity of the population structure was estimated by calculating a simple diversity index (SDI).

Results: One-hundred-and-twenty-one isolates (50.6%) produced carbapenemases, of which 87 (71.9%) were OXA-48, 27 (22.3%) were VIM-1, 4 (3.3%) were KPC-2, 2 (1.7%) were NDM and 1 (0.8%) was IMP-22; 4 isolates were collected in 2012, 40 in 2013 and 77 in 2014. Ertapenem was more sensitive than imipenem or meropenem for screening for OXA-48-producing *E. coli*. Using the Warwick typing scheme, 59 different STs were identified, the most prevalent being ST131 (16.5%). The population diversity was higher among VIM-1-producing isolates (SDI=81.5%) than among OXA-48-producing isolates (SDI=44.8%). The Pasteur scheme had a higher discrimination capability (SDI=55.4%) than the Warwick scheme (SDI=48.8%).

Conclusions: A progressive increase in the prevalence of CP *E. coli* was observed, mainly due to the dissemination of OXA-48 producers. The most sensitive method for detecting decreased susceptibility of CP *E. coli* to carbapenems was disc diffusion with ertapenem using the EUCAST screening cut-offs. The spread of CP *E. coli* was due to a polyclonal population. The Pasteur scheme showed the highest discrimination power. Surveillance is crucial for the early detection of CP *E. coli*.

Introduction

The increase in the number of carbapenemase-producing (CP) Enterobacteriaceae (CPE) is a serious threat to public health. This is because carbapenems are often the treatment option of last resort against infections caused by MDR Gram-negative bacilli.[1] Carbapenemases are β-lactamases that are able to hydrolyse nearly all the β-lactam antibiotics. The most clinically important carbapenemases in Enterobacteriaceae are the class A KPC, the class B VIM, IMP and NDM, and the class D OXA-48 type.[1] Thus far, the greatest epidemiological and clinical impact of CPE worldwide has been due to the high incidence of nosocomial infections caused by *Klebsiella pneumoniae*. However, over the last few years other CP species, such as *Enterobacter* spp., *Klebsiella oxytoca* and *Serratia marcescens*, have also been detected in nosocomial settings.[2]

In contrast to *K. pneumoniae*, much less is known about the CP *E. coli*. The information that is available is in reference to the NDM and OXA-48 classes.[2–4] Of concern is the fact that the acquisition of carbapenemases by successful *E. coli* clones, such as ST131, has already occurred.[4]

According to recent data,[5] eight phylogroups are now recognized, of which seven (A, B1, B2, C, D, E and F) belong to *E. coli sensu stricto* and one belongs to the *Escherichia* cryptic clade I. Recently, we observed that some highly virulent phylogroup A isolates could be further reclassified as phylogroup C according to this new classification.[6] Classically, most extraintestinal pathogenic *E. coli* isolates belong to phylogenetic group B2 and, to a

lesser extent, to group D. However, *E. coli* isolates of phylogroups A and B1 are mainly found as part of the intestinal commensal population.

MLST is an unambiguous method for characterizing bacterial population structure using the sequences of internal fragments of housekeeping genes. Currently there are two accepted *E. coli* MLST schemes: the University of Warwick scheme and the Institut Pasteur scheme.

The possible widespread occurrence of the CP *E. coli* could lead to a new epidemiological crisis similar to that seen with ESBL.[7] In this scenario, a likely fast clonal and polyclonal dissemination in a variety of settings, including community settings, would greatly increase the threat of CPE. In the light of this possibility, the aims of this study were: (i) to study the epidemiological and microbiological trends in CP *E. coli* isolates collected in Spain between 2012 and 2014; (ii) to analyse the population structure of CP *E. coli* in Spain using the two different MLST schemes; and (iii) to test and compare the discriminatory power of these MLST schemes.

Materials and methods

Study design and bacterial isolates

Since 2009, our official public health institute has been operating an active, unrestricted and non-mandatory National Antibiotic Resistance Surveillance Programme.[8] All carbapenem-non-susceptible *E. coli* detected between January 2012 and December 2014 were included in this study. Only the first isolate taken from each patient was analysed.

Standard microbiological methods and the API 20E system (bioMérieux, Marcy-l'Étoile, France) were used to identify the *E. coli* isolates. When necessary, the species identification was confirmed by 16S ribosomal DNA sequencing.

Antibiotic susceptibility and phenotypic characterization of carbapenemase production

Antibiotic susceptibility tests were performed using the disc diffusion method according to the EUCAST guidelines.[9,10] In addition, susceptibility to imipenem, meropenem, ertapenem, colistin, fosfomycin, tigecycline and temocillin was tested using MIC strips (Liofilchem, Italy). Carbapenem non-susceptibility was defined as decreased susceptibility to at least to one of the carbapenems tested. EUCAST screening cut-off values were employed in the analysis.[11]

Following EUCAST recommendations, inhibition of carbapenemase activity was carried out using EDTA, phenyl-boronic acid and cloxacillin.[11] ESBL production was detected using the combination disc and double-disc synergy test, according to EUCAST guidelines.[11] Carba NP method and the modified Hodge test with a meropenem disc containing 600 μg of cloxacillin[12,13] were carried out in all isolates.

Characterization of β-lactamases

The presence of genes encoding carbapenemases (bla_{OXA-48}, bla_{KPC}, bla_{VIM}, bla_{IMP} and bla_{NDM}),[8,14,15] ESBLs (bla_{TEM}, bla_{SHV} and bla_{CTX-M})[16] and plasmid-mediated AmpC (bla_{CMY}, bla_{FOX}, bla_{MOX}, bla_{DHA}, bla_{EBC} and bla_{ACC})[17] was determined using PCR and DNA sequencing assays. The sequences obtained were compared with those available in the publicly accessible GenBank and Lahey Clinic (http://www.lahey.org/Studies/) databases.

Molecular epidemiology

The phylogenetic groups were determined by PCR according to the new Clermont method.[5] MLST of each isolate was carried out using the Warwick (http://mlst.warwick.ac.uk/mlst/dbs/Ecoli) and Pasteur (http://bigsdb.web.pasteur.fr/ecoli/) typing schemes. Serotypes O25b and O16 were identified by allele-specific PCR, as described.[18,19]

The genetic relationship between the CP *E. coli* isolates belonging to the same ST was elucidated by PFGE after total chromosomal DNA digestion with XbaI.[16]

As previously described by Gastmeier *et al*.,[20] in order to analyse the population diversity a simple diversity index (SDI) was calculated as follows: number of STs/total number of isolates×100.

Statistical analysis

The significance of the CP *E. coli* trends was calculated using the χ^2 test for trends. Differences in the prevalence values for the resistance mechanisms of the different CP isolates and STs were assessed using Fisher's exact test. The null hypothesis was rejected when a P value ≤ 0.05 was calculated. Statistical analysis was performed using the GraphPad Prism software, version 3.02 (GraphPad Software, Inc., San Diego, CA, USA).

Results and discussion

Bacterial isolates

From January 2012 to December 2014, 239 isolates of carbapenem-non-susceptible *E. coli* were analysed. Of these, 121 (50.6%) produced carbapenemases. Over the same period of time, our reference laboratory identified a total of 2443 CPE, of which 121 (4.9%) were CP *E. coli*. A clear upward trend was observed in the percentage of CPE isolates identified as CP *E. coli* annually: 1.7% in 2012 (4 out of 237), 5.2% in 2013 (40 out of 775) and 5.4% in 2014 (77 out of 1431) (χ^2 test for trend=4.27; P=0.038). These CP *E. coli* isolates came from 39 Spanish hospitals, which were located in 12 geographical areas (range of isolates per hospital 1–16).

Sixty-six out of the total of 121 CP *E. coli* isolates (54.5%) were from males while 85 (70.2%) were from patients >65 years of age. Fifty-six out of the total number of CP *E. coli* isolates (46.3%) produced clinical infections, mainly urinary tract (20; 35.7%) or wound (20; 35.7%) infections. The remaining 65 isolates (53.7%) were obtained from carriers, mainly from rectal samples (n=61; 93.8%).

An upward trend in the detection of CP *E. coli* had been observed in other countries,[21,22] although the total number of cases remained low.[22–25] Of the 47 843 isolates of *E. coli* collected worldwide and studied by Peirano *et al*.,[4] only 116 (0.24%) were CP. According to the EARS-Net database, carbapenem resistance in blood isolates of *E. coli* is <1% in the vast majority of European countries.

The CP *E. coli* trend described in this study in Spain occurred after a continuous increase in resistance to third-generation cephalosporins (from 2.2% in 2002 to 12.5% in 2014) and fluoroquinolones (from 19.3% in 2002 to 34% in 2014) in blood isolates of *E. coli* reported to the EARS-Net database by Spanish hospitals (http://ecdc.europa.eu/en/healthtopics/antimicrobial_resistance/database/Pages/database.aspx). Furthermore, the overall prevalence of infection caused by CP *K. pneumoniae* increased from 0.2% in 2009 to 1.7% in 2013 in Spain.[2]

In a recent Spanish study,[26] 40% of infections caused by CPE, specifically OXA-48 producers, were community onset, but most of them were healthcare associated; only 9 of the 245 (3.6%) cases were community acquired.

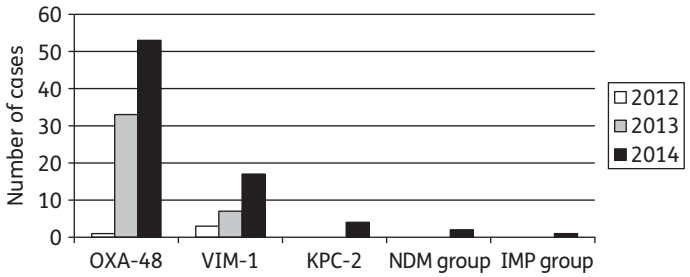

Figure 1. Annual evolution (2012–14) of CP *E. coli* in Spain.

Types of carbapenemases

Of the 121 CP *E. coli* isolates, 87 (71.9%) produced OXA-48; 27 (22.3%) produced VIM-1, 4 (3.3%) produced KPC-2, 2 (1.7%) produced NDM (1 NDM-1 and 1 NDM-5) and 1 (0.8%) produced IMP-22. Annual changes in CP *E. coli* number of cases according to the type of carbapenemase detected are shown in Figure 1. Consistent with the results of studies carried out in other countries,[22,24,27–31] the type of carbapenemase most frequently identified in *E. coli* in this study was OXA-48. KPC-producing Enterobacteriaceae are the main cause of the endemic CPE situation in some European countries, such as Italy and Greece.[27] However, the epidemiology of CPE has worsened in Europe over the last 2 years due to the rapid spread of *K. pneumoniae* producing OXA-48 in countries such as France, Spain and Turkey.[2,22,27,28] In contrast, in a worldwide study of ST131 CP *E. coli* carried out from 2008 to 2013 the carbapenemases most frequently identified were NDM followed by KPC.[4] The most prevalent *E. coli* carbapenemases in Portugal and China were KPC-3 and KPC-2, respectively.[23,32] Interspecies transmission of the bla_{OXA-48} gene is possible and has been shown to occur between *K. pneumoniae* and *E. coli*.[33]

Co-production of ESBL and plasmid-mediated AmpC enzymes

Thirty-seven out of the 121 (30.6%) CP *E. coli* isolates also produced ESBL. Of these, 16 (43.3%) produced CTX-M-15, 13 (35.1%) produced CTX-M-14, 6 (16.2%) produced SHV-12, 1 (2.7%) produced CTX-M-1 and 1 (2.7%) produced CTX-M-32. Production of ESBL was more frequently observed in the OXA-48-type isolates; ESBL was detected in 32 out of 87 (36.8%) of these isolates compared with only 3 of 27 (11.1%) VIM-1 isolates ($P=0.015$). In addition, 6 out of 121 (5%) CP *E. coli* isolates co-produced CMY-2; of these, 5 were OXA-48 producers and 1 was a VIM-1 producer.

The percentage of *E. coli* isolates co-producing OXA-48 and ESBLs was much lower than that reported for OXA-48-producing *K. pneumoniae* isolates.[2,34] In contrast with previous reports, we did not detect the CTX-M-24 ESBL, which is thought to associate with OXA-48-producing *E. coli*.[29,34,35]

Antibiotic susceptibility of CP E. coli

All 121 CP *E. coli* isolates were susceptible to tigecycline and colistin; 97.5% were susceptible to amikacin, 92.6% to fosfomycin, 74.4% to gentamicin, 58.7% to aztreonam, 55.4% to tobramycin, 38% to trimethoprim/sulfamethoxazole, 31.4% to ciprofloxacin and 20.7% to cefotaxime.

Of the 50 OXA-48 producers without resistance mechanisms to third-generation cephalosporins, 25 were intermediate or resistant ($n=23$) or highly resistant ($n=2$) to cefotaxime. It has been demonstrated that OXA-48 carbapenemase does not significantly hydrolyse ceftazidime and cefepime but hydrolyses cefotaxime very poorly.[28] The OXA-163 carbapenemase is a variant of OXA-48 that hydrolyses expanded-spectrum cephalosporins but has little hydrolytic activity against carbapenems.[36] This β-lactamase was not detected in the current study.

Laboratory detection of decreased susceptibility to carbapenems in CP E. coli

Using the EUCAST disc diffusion clinical breakpoints,[10] OXA-48- and VIM-1-producing isolates showed a higher percentage of decreased susceptibility to ertapenem (100% and 88.9%, respectively) than imipenem (35.6% and 63%, respectively) and meropenem (49.4% and 40.7%) (Table 1). Using EUCAST MIC clinical breakpoints, ertapenem also showed the highest percentage of non-susceptibility in OXA-48 producers (89.7%), but in VIM-1 producers imipenem had the highest non-susceptibility percentage (40.7%) (Table 1).

Using EUCAST screening cut-offs for carbapenemases,[11] OXA-48-producing isolates were best detected by ertapenem rather than meropenem or imipenem. This was true both when using the disc diffusion method and when using MIC strips (Table 1). VIM-1 producers were best detected by ertapenem when using disc diffusion and most effectively detected by meropenem when using MIC strips (Table 1). The specificity of the data was not calculated.

In general, disc diffusion was better for detecting the non-susceptibility of CP *E. coli* to carbapenems than MIC strips. In addition, employing ertapenem was better for screening OXA-48 *E. coli* than imipenem or meropenem. These observations were consistent with previous studies of OXA-48-producing Enterobacteriaceae.[30,34,37,38] However, using ertapenem as the only marker in the screening of carbapenemase activity in *Enterobacter* spp. should be avoided because of the many false positive cases observed.[2,11]

Temocillin has been proposed as a screening agent for OXA-48.[39] The MIC of this antibiotic was found to be >128 mg/L in 85 (97.7%) of OXA-48-producing isolates. The MIC of temocillin for the remaining two isolates was 4 and 8 mg/L, but they tested positive in the modified Hodge test. Woodford *et al.*[40] found that the MIC of temocillin was >128 mg/L in 87.9% of isolates of OXA-48-producing *E. coli*. This observation suggests that a high MIC of temocillin should not be the sole criterion for predicting the presence of OXA-48.

Laboratory detection of CP *E. coli* may be more difficult in comparison with CP *K. pneumoniae*, particularly in the case of OXA-48, because: (i) the isolates may appear susceptible to imipenem and meropenem (Table 1); and (ii) there is a high frequency (63.2% in this study) of OXA-48-producing isolates without ESBL co-production.

Phylogenetic groups

According to the new Clermont scheme,[5] 32 out of the 121 (26.5%) CP *E. coli* isolates studied belonged to phylogroup B2,

Table 1. Detection of non-susceptibility to carbapenems in CP *E. coli* using the methods of disc diffusion and MIC test strips, according to EUCAST clinical breakpoints and EUCAST screening cut-offs for carbapenemases

		Disc diffusion, *n* (%)			MIC test strips, *n* (%)		
Type of carbapenemase (*n*)	Carbapenem	S	NS	detection using EUCAST screening cut-offs for carbapenemases[a]	S	NS	detection using EUCAST screening cut-offs for carbapenemases[a]
OXA-48 (87)	ertapenem	0 (0)	87 (100)	87 (100)	9 (10.3)	78 (89.7)	86 (98.8)
	imipenem	56 (64.4)	31 (35.6)	47 (54)	56 (64.4)	31 (35.6)	57 (65.5)
	meropenem	44 (50.6)	43 (49.4)	74 (85.1)	75 (86.2)	12 (13.8)	84 (96.5)
VIM-1 (27)	ertapenem	3 (11.1)	24 (88.9)	24 (88.9)	22 (81.5)	5 (18.5)	15 (55.5)
	imipenem	10 (37)	17 (63)	19 (70.4)	16 (59.3)	11 (40.7)	18 (66.7)
	meropenem	16 (59.3)	11 (40.7)	19 (70.4)	26 (96.3)	1 (3.7)	20 (74.1)
KPC-2 (4)	ertapenem	0 (0)	4 (100)	4 (100)	0 (0)	4 (100)	4 (100)
	imipenem	0 (0)	4 (100)	4 (100)	0 (0)	4 (100)	4 (100)
	meropenem	0 (0)	4 (100)	4 (100)	0 (0)	4 (100)	4 (100)
NDM-like (2)	ertapenem	0 (0)	2 (100)	2 (100)	0 (0)	2 (100)	2 (100)
	imipenem	0 (0)	2 (100)	2 (100)	0 (0)	2 (100)	2 (100)
	meropenem	0 (0)	2 (100)	2 (100)	0 (0)	2 (100)	2 (100)
Total isolates (121)[b]	ertapenem	3 (2.5)	118 (97.5)	118 (97.5)	31 (25.6)	90 (74.4)	108 (89.3)
	imipenem	67 (55.4)	54 (44.6)	72 (59.5)	73 (60.3)	48 (39.7)	81 (66.9)
	meropenem	61 (50.4)	60 (49.6)	100 (82.6)	102 (84.3)	19 (15.7)	111 (91.7)

n, number of isolates; S, susceptible isolates; NS, non-susceptible isolates.
[a]According to Matuschek *et al*.[9]
[b]The single imipenem-producing isolate is also included in this analysis.

26 (21.5%) to phylogroup C, 21 (17.4%) to phylogroup B1, 16 (13.2%) to phylogroup E, 13 (10.7%) to phylogroup F, 12 (9.9%) to phylogroup A and 1 (0.8%) to phylogroup D.

Differences between the most frequent phylogroups of OXA-48- and VIM-1-producing isolates are shown in Table 2.

In contrast to previous reports, we observed a higher percentage of *E. coli* isolates in phylogroup C. This was because, using the new typing scheme, some clones were reclassified into this phylogroup, e.g. ST10 and ST23. By the classical Clermont scheme, both of these clones had previously been assigned to phylogroup A.[29] In a recent study, OXA-1-producing *E. coli* isolates were found to belong primarily to phylogroups C and B2.[6]

Population structure of CP E. coli isolates

Using the Warwick MLST scheme, 59 different STs were identified. The most frequent STs and their phylogenetic groups are detailed in Table 3.

ST131, which was the most common ST (20 isolates, 16.5% of the total isolates), was found in 13 of the 39 participating hospitals. These 13 hospitals were located in five different geographical areas. The ST131 isolate was most often found to carry the bla_{OXA-48} gene (14 of 20 isolates, 70%); however, bla_{VIM-1} was occasionally detected (4 isolates, 20%), as were KPC-2 (1 isolate, 5%) and NDM-5 (1 isolate, 5%). Out of the 20 ST131 isolates, 7 (35%) were ESBL producers; 6 of these produced CTX-M-15 and one produced SHV-12.

Table 2. Different population markers indicating the genetic variations between OXA-48- and VIM-1-producing *E. coli* isolates

	Resistance mechanism	
Population marker	OXA-48 (*N*=87)	VIM-1 (*N*=27)
Number of STs	39	22
Median of isolates per ST (range)	2.2 (1–14)	1.2 (1–4)
Number of single isolates by ST (%)	25 (28.7)	19 (70.4)
SDI, %	44.8	81.5
ST131, *n* (%)	14 (16.1)	4 (14.8)
ST68, *n* (%)	8 (9.2)	0
Phylogroup C, *n* (%)	22 (25.3)	3 (11.1)
Phylogroup B2, *n* (%)	21 (24.1)	8 (29.6)
Phylogroup B1, *n* (%)	13 (14.9)	6 (22.2)

Comparison between OXA-48- and VIM-1-producing isolates showed a more diverse population in VIM-1-producing isolates (Table 2).

Our results show that, in general, the population structure of CP *E. coli* is more polyclonal than that of CP *K. pneumoniae*.[2,41,42] *E. coli* ST131 is a high-risk clone that disseminates CTX-M-15 worldwide.[7] It is also most likely responsible for the global spread of bla_{KPC}-producing *E. coli*.[4] ST68 was previously described as a $bla_{CTX-M-14}$ carrier.[43] We did not detect ST38, a high-risk clone that had been associated with OXA-48-producing *E. coli* isolates.[24,34,35,37,38]

Table 3. Correspondence between the most prevalent STs obtained using the Warwick MLST scheme and the Pasteur MLST scheme; phylogroups of these primary STs are also included

Correspondence between the most prevalent Warwick STs and the Pasteur STs			Correspondence between the most prevalent Pasteur STs and the Warwick STs		
phylogroup	Warwick ST (*n*)	Pasteur ST (*n*)[a]	phylogroup	Pasteur ST (*n*)	Warwick ST (*n*)[b]
B2	131 (20)	9 (10), 43 (8), 506 (1), 729 (1)	B2	9 (10)	131
E	68 (8)	14 (7), 665 (1)	B2	43 (9)	131 (8), 2279 (1)
C	10 (7)	2 (4), 478 (2), 680 (1)	C	2 (8)	10 (4), 44 (1), 167 (1), 744 (2)
C	23 (5)	7 (5)	E	14 (7)	68 (7)
C	410 (4)	471 (4)	C	7 (5)	23 (5)
F	648 (4)	662 (2), 721 (2)	B1	88 (5)	101 (2), 359 (2), 3024 (1)
B1	1431 (4)	649 (4)	C	471 (4)	410 (4)
			B1	649 (4)	1431 (4)

n, number of isolates.
[a]All Pasteur STs belonging to a single Warwick ST were either a single-locus variant or a double-locus variant, except for ST506, which had five different alleles related to ST9, ST43 and ST729.
[b]All Warwick STs belonging to a single Pasteur ST were either a single-locus variant or a double-locus variant.

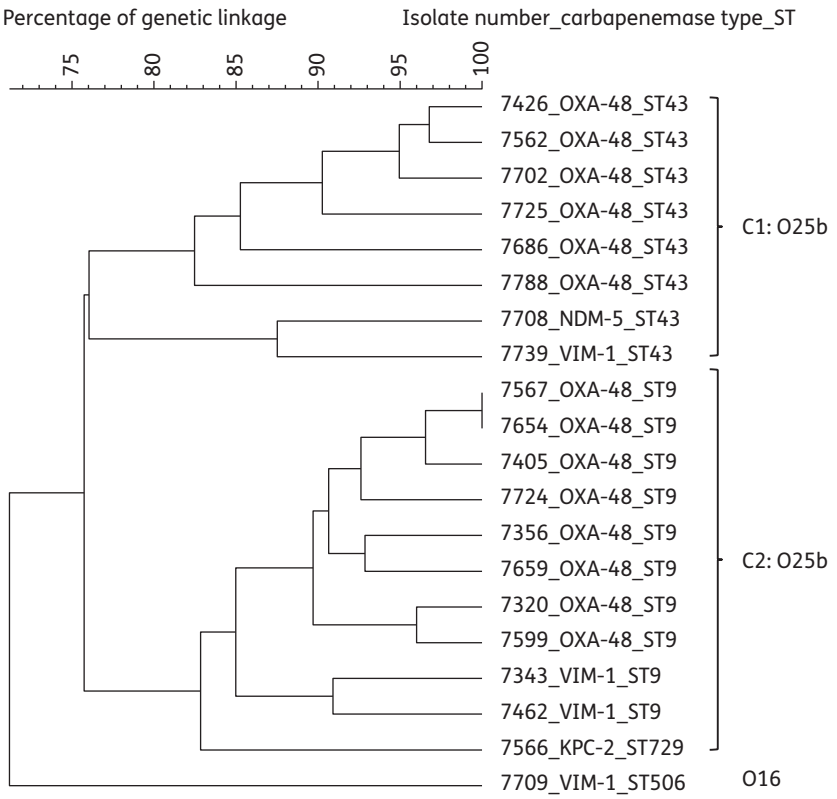

Figure 2. Dendrogram illustrating the genetic relationships between 20 CP *E. coli* isolates belonging to the Warwick-derived ST131. Isolate numbers, carbapenemase types and Pasteur-derived STs are arranged to the right of the dendrogram.

Comparison of MLST by the Pasteur and Warwick typing schemes

Sixty-seven different STs were identified using the Pasteur typing scheme. The most prevalent STs and their phylogenetic groups are included in Table 3. There was a non-univocal correlation between the STs determined by the two schemes. Some of the more prevalent STs identified by the classic Warwick scheme corresponded to several STs identified by the Pasteur scheme and vice versa (Table 3). The predominant ST131 clone identified with the

Warwick scheme corresponded to four different STs identified with the Pasteur scheme; these were ST9, ST43, ST506 and the newly identified ST729 clone. The correspondence between the Warwick-derived ST131 and the Pasteur-derived ST43 and ST506 had been described previously.[44] These data suggest that, within a single ST, additional population diversity could exist. However, correspondence between phylogroups and STs obtained by both MLST schemes was constant (Table 3). The ability to identify clones/phylogroups in *E. coli* is crucial, as a number of ecological and clinical features vary with its phylogenetic origins.[45]

This study provides novel and valuable information about the population structure, determined by two different MLST schemes, of an extensive collection of CP *E. coli* isolates. The Pasteur MLST scheme had a higher discrimination capability (SDI = 55.4%) than the Warwick MLST scheme (SDI = 48.8%).

PFGE assays showed that all isolates belonging to the same Warwick-derived ST had a genetic homology of >75% (data not shown). However, in the case of the Warwick-derived ST131, two main clusters, C1 and C2, were identified by PFGE. These clusters matched three Pasteur STs: ST43, which contained 8 isolates and corresponded to C1; ST9, which had 10 isolates and corresponded to C2; and ST729, which contained 1 isolate and corresponded to C2 (Figure 2). These 19 isolates corresponding to C1 and C2 belonged to the serotype O25b (Figure 2). The remaining ST131 isolate was not related to clusters C1 and C2, corresponded to the Pasteur-derived ST506 and belonged to the serotype O16 (Figure 2). Previously, O16-ST131 isolates have been described in the USA, Europe and Asia as a phylogenetic clade distinct from the classic O25b-ST131 clone.[44]

According to the typing data (phylogroups, MLST and PFGE) provided in this study, the population structure of CP *E. coli* in Spain appears to be very diverse (Tables 2 and 3), suggesting that plasmid-mediated transmission of carbapenemases may be highly effective.

Conclusions

In summary, we carried out a multicentre study in order to characterize the microbiological and molecular determinants of CP *E. coli* in Spain. Although the overall ability to produce carbapenemase remained rare in *E. coli*, an important upward trend was observed over the last few years, mainly due to the proliferation of OXA-48 producers. The disc diffusion method with ertapenem according to EUCAST criteria was the best method for detecting the reduced susceptibility to carbapenems in CP *E. coli*. A polyclonal population of CP *E. coli* was observed. The high-risk Warwick-derived ST131 clone predominated. The Pasteur typing scheme had a higher discrimination capability than the Warwick scheme. The classical Warwick-derived ST131 clone corresponded to four different Pasteur-derived STs: ST9, ST43, ST506 and ST729. The early identification of CP *E. coli*, including detection of the high-risk clones implicated in their spread, and the quick and suitable implementation of infection control measures, are crucial for limiting their dissemination in both the hospital and the community setting. Expert guidelines on infection control measures for CPE have been provided by the European Society of Clinical Microbiology and Infectious Diseases.[46]

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Transparency declarations

None to declare.

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